
**REPORT TO THE SENATE ARMED SERVICES
COMMITTEE AND THE HOUSE
ARMED SERVICES COMMITTEE**

on

**Department of Defense
Animal Care and Use Programs 1998**

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LIST OF ACRONYMS

AAALAC	Association for Assessment and Accreditation of Laboratory Animal Care International
AALAS	American Association of Laboratory Animal Science
ACLAM	American College of Laboratory Animal Medicine
ADCNO (MP&T)	Assistant Deputy Chief of Naval Operations for Manpower, Personnel, and Training
ADCS (M&RA)	Assistant Deputy Chief of Staff for Manpower and Reserve Affairs
ADCSPER	Army Assistant Deputy Chief of Staff for Personnel
AL/HR	Armstrong Laboratory/Human Resources
APHIS	Animal and Plant Health Inspection Service
ARI	Army Research Institute
ASBREM	Armed Services Biomedical Research Evaluation and Management
AWA	Animal Welfare Act
AWIC	Animal Welfare Information Center
BRD	Biomedical Research Database
CDR, AFHSD	Commander, Air Force Human Systems Division
CNS	Central Nervous System
CRISP	Computer Retrieval of Information on Scientific Projects
DDR&E	Director, Defense Research and Engineering
DoD	Department of Defense
DTIC	Defense Technical Information Center
FDA	Food and Drug Administration
FEDRIP	Federal Research in Progress
FY	Fiscal Year
GME	Graduate Medical Education
IACUC	Institutional Animal Care and Use Committee
IG	Inspector General
ILAR	Institute of Laboratory Animal Research
IRAG	Interagency Regulatory Alternatives Group
JDL	Joint Directors of Laboratories
JTCG	Joint Technology Coordinating Groups
LAM	Laboratory Animal Medicine
MATRIS	Manpower and Training Research Information Services
NIH	National Institutes of Health
NMR	Nuclear Magnetic Resonance
NOS	Nitric Oxide Synthase
NPRDC	Navy Personnel Research and Development Center
NRC	National Research Council
NTSC	Naval Training Systems Center
OPRR	Office for the Protection from Research Risks
OSD	Office of the Secretary of Defense

PALS	Pediatric Advance Life Support
PCR	Polymerase Chain Reaction
PHS	Public Health Service
PL	Public Law
POC	Point of Contact (Primary Contact)
RDT&E	Research, Development, Test, and Evaluation
S&T	Science and Technology
STO	Science and Technology Objective
STRICOM	Simulation Training and Instrumentation Command
TAPSTEM	Training and Personnel Systems Science and Technology Evaluation and Management
USAMRMC	United States Army Medical Research and Materiel Command
USDA	United States Department of Agriculture
VEE	Venezuelan Equine Encephalitis
WRAIR	Walter Reed Army Institute of Research

SECTION I

INTRODUCTION/OVERVIEW

This is the Fiscal Year (FY) 1998 Report to Congress on Department of Defense Animal Care and Use Programs. In addition to a general overview, this report provides a detailed account of Department of Defense (DoD) animal use; to include its publicly accessible database, animal care and use oversight procedures, Institutional Animal Care and Use Committees (IACUCs), alternatives to animal use programs, Association for Assessment and Accreditation of Laboratory Animal Care International (AAALAC) status, and animal use.

The report covers animal research conducted by the DoD including education, training, and testing both in DoD laboratories and by extramural projects funded by the Department for FY98. This report does not include information on animals used by the DoD solely for the purpose of food preparation for human or animal consumption, ceremonial activities, recreation, or the training, care, and use of military working animals.

I.1 REQUIREMENTS FOR USE OF ANIMALS IN THE DoD

DoD use of animals in research, development, education, and training is critical to sustained technological superiority in military operations in defense of our national interests. The DoD's biomedical research, development, test, and evaluation (RDT&E) training programs that are dependent upon animal use ultimately translate into improved military readiness as well as reduction in morbidity and mortality associated with military operations. These programs contribute directly to ensuring that service men and women maximize their capabilities to survive the numerous and various hazards they face around the world.

DoD research has benefited greatly from animal use alternatives such as non-living systems, cell and tissue culture, and computer technology. However, complex human organ systems interactions, in addition to environmental factors and confounding variables, necessitate the continued judicious use

of animal models in DoD programs. Although many innovative animal use alternatives have been developed and are in use by Department scientists, situations remain in which there are no acceptable non-animal alternatives available. For example, there are no adequate models addressing the movement and general effects of drugs, toxicants, or pathogens in the body. Similarly, cell and tissue cultures are limited in their abilities to simulate endocrine, neurological, immune, or inflammatory responses. As new advances, technologies, and breakthroughs in animal use alternatives occur, the DoD will embrace them whenever possible. The chapter on alternatives in this report gives a full account of the aggressive programs and numerous animal use alternatives implemented in DoD laboratories.

Disease remains a major cause of death and disability in military operations and conflicts. During Operations Desert Storm and Restore Hope, outbreaks of respiratory diseases, diarrheal diseases such as shigellosis, and parasitic diseases such as leishmaniasis and malaria, threatened the health and well-being of our troops. Indeed, the DoD is still assessing and addressing concerns over the long-term effects of various environmental, physical, and medical factors associated with the Persian Gulf Conflict. It is obvious that the health and well-being of military personnel extend far beyond the immediate scope of the battlefield. We have an irrefutable moral obligation to our soldiers, sailors, airmen, and marines to provide the maximum protection and care possible. DoD researchers are committed to accomplishing this goal, and in many cases, animal-based research is the critical underpinning for the fulfillment of that obligation.

One of the most critical areas requiring DoD animal use is the compelling need to develop vaccines, drugs, and therapies to protect, sustain, and treat service men and women during military operations. These research programs are strongly focused on a myriad of militarily relevant diseases and threats, many of which can result in potentially fatal diseases or conditions that have no known

treatments, therapies, or cures. Consequently, there are numerous instances, including medical chemical and biological warfare defense, where animal-based studies are particularly critical. Ethical concerns, as well as regulatory requirements of the Food and Drug Administration (FDA), necessitate that candidate vaccines and drugs be safe and efficacious in laboratory animal models prior to initiation of human use protocols whenever possible. The rationale for this is to prevent the fielding and use of ineffective or dangerous treatments. Indeed, during the final stages of vaccine and drug development, large-scale safety and efficacy testing is usually conducted using human volunteers. However, in the search for understanding and developing protection against many highly lethal agents, human use protocols are simply not possible. Consequently, carefully regulated animal use is absolutely vital to the success of Department biomedical research programs. The ultimate goal is to maximize the survivability of our troops in all situations.

Additionally, many examples of the humanitarian benefits of the DoD investment in animal research that are shared on an international basis improve the quality of life of both humans and animals. Several prime examples of the humanitarian benefits of DoD research efforts are: the Junin vaccine that has provided critical protection for more than 120,000 individuals in endemic areas of Argentina against the ravages of Argentinean hemorrhagic fever; DoD-developed Venezuelan equine encephalitis (VEE), eastern equine encephalitis, and western equine encephalitis vaccines that have been used to limit and control epidemics of VEE in Venezuela and Colombia in 1995, and to protect occupational workers in vaccine production plants around the world. In addition to being important public health tools, the equine encephalitis vaccines are obviously critical adjuncts to animal health programs around the world.

The DoD must develop the materiel and technological means to best protect and sustain the health and well-being of service men and women against all threats, and provide the best medical treatment possible to those who become casualties. This responsibility underlies the need for the DoD to conduct research, and to train and educate military health care providers in the most effective medical management of battlefield casualties. Battlefield health care must very often be provided

in an austere, harsh, and hostile environment, hours away from a definitive care hospital, unlike medical counterparts found in civilian emergency medicine and trauma management. A domestic, low velocity projectile gunshot patient in a modern civilian shock and trauma center will be supported and resuscitated by a full complement of medical staff with a plentiful supply of oxygen, fluids, medications, surgical intervention, and nursing. The combat casualty may be supported by only a single aidman and the medical supplies, experience, and expertise he can carry.

Clinical investigation programs at Medical Treatment Centers are provided to support Graduate Medical Education programs (GME). GME programs are post doctoral programs to train physicians in residency programs to specialize in pediatrics, orthopedics, surgery, etc. To be certified, the GME programs must demonstrate that the Medical Center has programs to provide research opportunities for both staff and students. The clinical investigation programs provide the training in research, protection of human subjects, and use of animals in research. They provide opportunities not only for staff and GME students, but for patients who desire to participate in research protocols, such as Multicenter Oncology and Pediatric Oncology protocols. In this regard, Congress has mandated that DoD will work closely with the National Institutes of Health to provide more opportunities for DoD beneficiaries to participate in National Institutes Health sponsored protocols. Many of the clinical investigation training protocols support GME programs, such as Advanced Trauma Life Support and Pediatric Advanced Life Support (PALS) courses which follow requirements set by the American College of Surgeons, and the training protocol using ferrets, cats or rabbits for intubation for pediatric intubation training. There may also be surgical skills training courses for micro, vascular, or reproductive surgery. Programs using animals for GME training have oversight of procedures by veterinarians and are conducted in AAALAC certified facilities.

The use of animals is important in the DoD's non-medical programs. These studies include the development of biological sensors, sonar, echolocation, biorobotics, aviation construction materials and hearing and eye protection systems. There are also non-medical studies to understand learning and memory physiology in an attempt to

model the brain's circuitry for advanced data processing computers and robotic machinery. These advanced computers and robots will eventually reduce the risk that our servicemen and women encounter in their daily duties. The DoD performs marine biology research to better understand the military working marine mammals. In addition, the marine mammals are investigated to determine their auditory detection thresholds in marine use as sentries. Studies of biosonar systems to enhance the use of military marine mammal systems for mine detection and retrieval, personnel detection, and reconnaissance.

I.2 DoD POLICY GOVERNING ANIMAL RESEARCH

The DoD is committed to full ethical and regulatory compliance for its animal-based research programs. The DoD has been proactive in increasing the fixed infrastructure and span of control necessary to ensure lawful and efficient execution of programs and maximize oversight of diverse and varied missions. The Department has aggressively implemented focused programs and working documents that optimize standardization of animal care and use at the user level. This enhanced standardization and oversight have improved a historically good system, and made it outstanding.

In 1995, the DoD revised and implemented the directive dealing specifically with animal care and use (DoD Directive 3216.1, "The Use of Animals in DoD Programs," 1995) (Appendix A). This directive strengthens and clarifies requirements for nonaffiliated membership on IACUCs and directs all DoD animal use facilities that maintain animals for research, testing, and training to apply for AAALAC accreditation.

The DoD also implemented a Policy Memorandum entitled "Department of Defense (DoD) Policy for Compliance with Federal Regulations and DoD Directives for the Care and Use of Laboratory Animals in DoD-Sponsored Programs" (Appendix B). This 1995 Policy Memorandum specifies training requirements for nonaffiliated DoD IACUC members and implements a standard format for animal use protocols (Appendix C), a standard checklist for IACUC inspections (Appendix D), and a standard

reporting requirement for all animal use research to support a publicly accessible database (Section II).

All animal research must conform to requirements of the 1966 Animal Welfare Act (Public Law [PL] 89-544) as amended in 1970 (PL 91-579), 1976 (PL 94-279), and 1985 (PL 99-198), as well as the National Research Council *Guide for the Care and Use of Laboratory Animals*, (7th rev. edition, 1996), U.S. Government Principles for Animal Use (1985) (Appendix E), and the requirements of the applicable regulations of the United States Department of Agriculture (USDA).

Although the Animal Welfare Act currently exempts mice and rats in the genera *Mus* and *Rattus*, the DoD has long afforded them, along with all other vertebrates, the same consideration given non-exempt species under the Animal Welfare Act. At the same time, DoD researchers have aggressively developed novel procedures to replace, reduce, and refine the use of animals during experimentation.

I.3 BENEFITS OF ANIMAL RESEARCH

DoD laboratories and extramural contractors provide the capability to solve the medical and non-medical problems of the future through the efforts of internationally renowned medical and scientific experts working in state-of-the-art facilities and in the field. The Department conducts or funds research, development, training, and evaluation to sustain the operational capabilities of today's service men and women. As noted in the previous section, many of these programs require the use of animals to meet their mission requirements. These programs result in many benefits for both the military and civilian sector (Table I-1). The military benefits from programs that do research in areas that currently threaten military personnel such as combat trauma, chemical and biological agents, infectious diseases not endemic to the United States, directed energy, and occupationally unique health hazards from military operations and environmental extremes. These research programs focus heavily on the prevention of casualties. They contribute significantly to the readiness and sustainment of the DoD's warfighting capability, and also to a significant reduction in the number of casualties reaching the medical treatment facilities.

Table I-1 *Animal Use Benefits*

<p>Medical</p> <p>Development of a vaccine for tick-borne encephalitis</p> <p>Monitoring the prevalence of a newly introduced dengue virus, dengue 2, in Peru</p> <p>Development of model with which to test DNA vaccines against malaria</p> <p>Identification of effective drugs and drug combinations for use as antiseizure nerve agent countermeasures</p> <p>Preparation and testing of candidate topical skin protectants against chemical agent exposure</p> <p>Creation of genetically engineered protective vaccine candidates for four types of staphylococcal enterotoxins</p> <p>Demonstration of the first effective vaccine candidate for the Marburg filovirus</p> <p>Study of hormone-related gender differences in thermo-regulation</p> <p>Refinement of a model of hemorrhagic hypotension with which to perform injury research</p> <p>Determination of the toxicity of tungsten, proposed to replace depleted uranium in munitions</p> <p>Assessment of the dermal absorption and cutaneous toxicity of topically exposed military jet fuels to skin</p> <p>U.S. patent obtained for use of substance P analog in highly effective treatment of melanoma cancers</p> <p>Vaccine development</p> <p>Research in breast, prostate, ovarian cancer</p> <p>Research on Gulf War Illnesses and neurofibromatosis</p> <p>FDA safety and efficacy testing prior to use in humans</p> <p>Development of medical countermeasures to military nerve and blister agents</p> <p>Prevention, diagnosis, and treatment of schistosomiasis and malaria</p> <p>Development of treatments to promote bone healing and growth</p> <p>Identification and monitoring of environmental toxins</p> <p>Clinical</p> <p>Development of a highly effective dry fibrin sealant bandage</p> <p>Development of expandable metal stents for treatment of tracheal injuries</p> <p>Development of methods to re-establish bone and bone growth in surgery and osteoporosis</p> <p>Improvement in patient care</p> <p>Non-Medical</p> <p>Assessment of the dermal absorption and cutaneous toxicity of topically exposed military jet fuels to skin</p> <p>Determination of toxic potential, or lack thereof, of contaminated ground and surface waters</p> <p>Development of biosensors</p> <p>Training</p> <p>Special forces medical training</p> <p>Advanced trauma life support training</p> <p>Graduate medical training</p>
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In addition, the DoD is involved in medical research that directly benefits the civilian population such as research in breast, prostate, and ovarian cancer, cardiovascular disease, trauma care and treatment, respiratory injuries, burns, and specific surgical procedures. A list of specific benefits by research category is shown in [Appendix F](#).

Besides the medical benefits of animal research, there are many non-medical and training benefits. The development of biosensors and the identification of environmental toxins benefit both the military and civilian communities. The DoD has many exceptional medical and scientific educational programs that train both medical personnel and scientists. While these people are in the military, the DoD reaps the benefit of this training; once they leave the military and apply their training in the private sector, this benefit is realized by the civilian community. The development of alternatives to animal use by the DoD provides an extra value to both communities and to animals as they discover ways to reduce or replace the use of animals. Also, refinement research results in more humane methods of performing research that is applied in many types of research settings.

In FY98, the DoD reported over 360 publications in scientific journals, proceedings, technical reports, books and book sections from RDT&E efforts using animals. Examples of both journal publications and proceedings by research category are presented in [Appendix G](#). In addition, this year the Navy reported receiving a patent (#77.468) for a recombinant dengue virus envelope protein/maltose-binding protein antigens and subunit vaccine compositions containing said antigens.

I.4 SCOPE OF REPORT

This report provides a comprehensive account of DoD animal care and use programs. There are sections that include in-depth discussions of:

- Publicly accessible information on Department research ([Section II](#));
- Policies and procedures for oversight of Department animal care and use programs ([Section III](#));
- AAALAC accreditation for Department animal care and use programs ([Section IV](#));

- d. DoD animal use profiles ([Section V](#)); and
- e. DoD initiatives to promote alternative methods that replace, reduce, or refine animal use ([Section VI](#)).

I.4.1 Publicly Accessible Information on Animal Use in the DoD

On October 1, 1995, the Department of Defense implemented a publicly accessible database analogous to the National Institutes of Health Computer Retrieval of Information on Scientific Projects System. The DoD Biomedical Research Database (BRD) is available on-line to the public, and is composed of succinct summaries of Department research projects, allowing interested individuals easy access to Department research information. The cost of animal-based research is presented by work unit summary in the BRD. In order to prevent duplication, this information is not presented in this report. More information on accessing the database is presented in [Section II](#).

I.4.2 Oversight of DoD Animal Care and Use Programs

DoD animal use oversight is reviewed in [Section III](#). In general, internal and external oversight provisions for animal research conducted by the DoD are at least as stringent as those for research in any other department of the federal government, and in many ways exceed the standards. As a matter of policy, the DoD abides by the applicable federal regulations pertaining to animal care and use, including provisions for oversight. All DoD facilities and extramural institutions sponsored by the DoD must submit proposals for animal use to an IACUC. The IACUCs review proposed animal protocols to ensure compliance with the Animal Welfare Act, and address concerns of the community. DoD Directive 3216.1 (1995) establishes oversight requirements that exceed the provisions of the Animal Welfare Act. Each IACUC serves as an independent decision-making body for the institution and establishes policy for the care and use of animals at that facility in accordance with applicable DoD directives, and federal law and regulations.

The DoD has developed and implemented a standardized protocol format for use by all of its units ([Appendix C](#)). It includes requirements for

searching the Federal Research in Progress database or an equivalent database and the Defense Technical Information Center database to prevent duplication of ongoing federally funded research. The principal investigator must justify the use of animals, including consideration of alternatives, justify the choice of species and the number of subjects, and include a literature search and assurance that the work does not needlessly duplicate prior experimentation. The protocol must specify procedures to be used with animals and methods to avoid or minimize pain. It must include a literature search for possible alternatives, qualifications of the individuals conducting procedures with animals, and disposition of animals at the termination of the work.

The IACUC ensures that personnel involved in animal-based studies are properly trained and, if necessary, establishes a training program to support the staff. The IACUC inspects facilities and animal care programs at least twice annually, and prepares a written report including a plan to address deficiencies. It enforces compliance with procedures specified in the protocols by conducting inspections, evaluating, and, if necessary, investigating reports of deviation from approved procedures. The IACUC of each facility performs semiannual program reviews of all animal use areas. The DoD 1995 Policy Memorandum ([Appendix B](#)) strengthens that process by establishing a standardized semiannual review checklist that outlines the areas required for IACUC review. This guidance is consistent with the recommendations of the DoD Inspector General (IG) report of February 1994 ([Appendix H](#)). A formal report of inspection shall be prepared twice annually, noting the use of the checklist, and indicating all major and minor deficiencies, a plan for correction of deficiencies, signatures of a majority of IACUC members, and a statement indicating whether there are or are not minority opinions. Finally, the IACUC serves as an impartial investigator of reports of violations of good animal practices and is empowered to suspend the use of animals for protocols not conducted in accordance with the Animal Welfare Act or institutional policy.

DoD Directive 3216.1 (revised in 1995) clarifies composition, membership, and training requirements of the IACUC. The 1995 modification addressed the House Armed Services Committee's request to improve community representation and

to appoint animal advocates to the Department's IACUCs, consistent with a recommendation of the IG Report of February 1994. The revised Directive (1995) increased the minimum membership of all DoD IACUCs from three to five. In addition, it specifies that

“there shall be at least one non-scientific member on the IACUC. In addition, there shall be at least one member representing the general community interest who is nonaffiliated with the research facility. The nonaffiliated member and the non-scientific membership can be filled by the same person. To ensure community representation at each meeting and inspection, an alternate to the nonaffiliated member shall be designated for all IACUCs having a single non-affiliated membership.”

Each DoD IACUC has increased its membership to comply with this Directive. Currently, about 25% of DoD IACUC members are non-scientific.

This Directive exceeds the requirements of the Animal Welfare Act and is further strengthened by the DoD 1995 Policy Memorandum, which requires a minimum of 8 hours of training for new non-affiliated members. In support of this training, the DoD developed a program consisting of a set of topics and recommended resources that may be used by individual IACUCs.

Responsibility for oversight of the Department's science and technology programs rests with the Director, Defense Research and Engineering (DDR&E). The staff, in conjunction with representatives from the Services, annually review the science and technology efforts to ensure they are fully coordinated and without unnecessary duplication of effort. The preponderance of animal use within the Department occurs in biomedical programs. These activities receive specific oversight from the Armed Services Biomedical Research Evaluation and Management (ASBREM) Committee, which was created by congressional direction in 1981. The ASBREM Committee is chaired by the DDR&E and co-chaired by the

Assistant Secretary of Defense (Health Affairs). The overall biomedical effort is carefully integrated and reviewed to eliminate unjustified duplication of effort by six subordinate Joint Technology Coordinating Groups reporting to the co-chairpersons.

I.4.3 Accreditation of DoD Laboratories by AAALAC

Animal use programs in the DoD strive to meet all the requirements of AAALAC. AAALAC accreditation is recognized as the “Gold Standard” for animal care and use programs. DoD Directive 3216.1 (1995) states that all DoD laboratories that maintain animals for use in research, testing, or training shall apply for AAALAC accreditation. Currently there are 36 DoD animal facilities worldwide. Of these, 33 (92%) were accredited in FY98.

Over the past 6 years, the DoD has been resolute in pursuing AAALAC accreditation for all of the facilities that use animals in research. This diligence has resulted in an increase in accreditation from 60% in FY93 to 92% in FY98.

I.4.4 DoD Animal Use Profiles by Research Category

A profile of DoD animal use is provided in [Section V](#). In this report, a detailed system was adopted for classifying animal use that includes 8 categories with 23 subcategories: 8 medical research, 4 non-medical research, 3 clinical research, 2 training, and 6 other categories of studies and use. Detailed charts and graphs are included in [Section V](#).

In FY98, the DoD used 291,551 animals, which is an 8% decrease from FY97. Of these, 26,750 (9%) were USDA reportable species as defined in the Animal Welfare Act of 1985. [Table I-2](#) summarizes the major animal use statistics for DoD research.

In addition, it should be noted that no animals were reported as used for development or testing of offensive weapons. During the time that the DoD has been reporting animal use to Congress (FY93-FY98), there has been a 47% decrease in the total number of animals used.

Table I-2 Summary of DoD Animal Use Statistics

Total Animal Use by Species	% of Total
Rodents, fish, amphibians, reptiles, and birds	96.18
Rabbits	1.28
Farm animals (i.e., sheep, pigs, cows, horses, goats, and burros)	1.72
Dogs, cats, nonhuman primates, marine mammals	0.71
Other	0.11

Percentages may not add up to 100% due to rounding of calculations.

Total Animal Use by Category	% of Use
Medical RDT&E	81.87
Non-Medical RDT&E	4.26
Clinical Investigation	3.92
Adjuncts/Alternatives	5.78
Training & Instructional	1.66
Breeding Stock	1.32
Classified Secret or Above	0.09
Other	1.09

Percentages may not add up to 100% due to rounding of calculations.

1.4.5 DoD Initiatives to Promote Alternative Methods that Replace, Reduce, and Refine the Use of Animals

Congress requested that the DoD establish aggressive programs to replace, reduce, and refine current use of animals. A review of DoD programs and initiatives to develop and implement alternatives to animal research is reviewed in [Section VI](#). Alternatives presented are those developed by DoD investigators and the general and specific alternatives implemented by the DoD in FY98.

Animal research is an essential part of the scientific process, but it is only initiated after due consideration of alternatives. The DoD uses a Standard Protocol Format that specifically requires each investigator to consider alternatives to the use of animals and to justify the animal model selected.

The IACUC process also includes a strong emphasis on consideration of alternatives in all protocols. In addition, all protocols that involve unrelieved pain or discomfort require consultation with a veterinarian prior to IACUC review, and a specific database search for scientifically acceptable alternatives to the proposed method. Each protocol that involves animals in research or training must explain the need for the animal research and defend the choice of species as the most scientifically valid model. Often, economies of time and resources are gained when scientifically valid alternatives to animal use are available. Our review of current animal research reveals that scientists in the DoD have developed or adopted many alternative methods based on ethical considerations and other inherent benefits. [Table I-3](#) presents examples of

Table I-3 Examples of Alternatives for Replacement, Reduction, and Refinement of the Animals Developed or Being Developed by the DoD

- DoD investigators have developed an artificial eye with lenses that can mimic the focusing characteristics of the eye.
- Realistic biophysical models computationally simulate the damage processes induced by lasers as accurately as possible.
- Artificial retina to be exposed to ultra short laser pulses to determine trends in damage threshold as a function of pulse width, wavelength, and number of pulses has been developed.
- In estrogen receptor research, methods have been developed to harvest and store uterine tissues at -80°C for several weeks, resulting in maximal use of the animals.
- Detailed postoperative discomfort monitoring and extended analgesic have been developed to decrease pain and distress.
- A reproductive toxicity test to replace laboratory mammals has been developed using the gametes and embryos of *Xenopus laevis*.
- A virtual reality simulation of surgical training has been developed.

alternatives developed by the Department in FY98 to replace, reduce, and refine the use of animals. In addition, the Department sponsors conferences and workshops to promote alternatives to animal research.

In FY98, over 500 animal use projects reported that they were implementing alternative methods to the use of animals. They implemented both general and specific alternatives. General alternatives are those that are frequently implemented in many different DoD programs. Specific alternatives are those that may be specific to both a research protocol and/or facility.

The DoD has funded the Institute of Laboratory Animal Research (ILAR) of the National Research Council to develop institutional training materials, education, and publications in support of DoD laboratory animal care and use programs since 1987. The Department has resolved to maintain this important collaboration by providing in excess of \$125,000 annually for the ILAR Program.

In conclusion, because the use of animals in research is essential to protect the health and lives of military personnel, the DoD must conduct research involving the use of animals for the foreseeable future. While research has benefited greatly from animal use alternatives, the confounding variables imposed by the complex interactions of organ, tissue, cell, and environmental factors necessitate the continued, judicious use of animal models in DoD programs. Animals are used in research only when scientifically acceptable alternatives are not available. The DoD is committed to full ethical and regulatory compliance for its animal-based research programs, and its animal care and use requirements are as strict or stricter than those required of non-DoD, government-funded, public and private research institutions. DoD policy directs all facilities maintaining animals for use in research, testing, or training to apply for AAALAC accreditation, and the DoD has established programs to replace, reduce, and refine current use of animals.

SECTION II

PUBLICLY ACCESSIBLE INFORMATION ON ANIMAL USE IN THE DoD

II.1 CONGRESSIONAL REQUEST INFORMATION

House Armed Services Committee Report 4301 (1995) requested the Secretary of Defense to “develop a mechanism for providing Congress and interested constituents with timely information... about [Department of Defense (DoD)] animal use programs, projects and activities, both intramural and extramural.” In response to this request, and to serve the interest of both the scientific community and general public, the Department has implemented a publicly accessible database called the Department of Defense Biomedical Research Database (BRD). The BRD is a database containing succinct summaries of the Department’s research projects involving the use of animals. This database is analogous to the National Institutes of Health (NIH) Computer Retrieval of Information on Scientific Projects (CRISP) System. The CRISP System is a biomedical database containing information on research projects supported by the United States Public Health Service, as well as information on intramural research programs of the NIH and the Food and Drug Administration. The BRD became accessible to the public through the Internet on October 1, 1995. It is located on the Manpower and Training Research Information Services (MATRIS) home page.

II.2 THE FY97 BIOMEDICAL RESEARCH DATABASE

The data in the FY97 BRD were developed from the current work unit summary system of the Defense Technical Information Center (DTIC). DoD organizations performing research, development, test, and evaluation (RDT&E) projects are currently mandated to provide annual reports of research to the DTIC. The DTIC maintains these work unit summaries in a database. While the majority of DoD animal use occurs in RDT&E projects, some work is performed in clinical investigations

programs that are not mandated to provide work unit summaries to the DTIC. Therefore, the DoD directed that these non-RDT&E DoD animal research projects develop summaries to be entered into the BRD. The areas of research, testing, and training in the FY97 BRD include, but are not limited to, the following: infectious diseases, biological hazards, toxicology, medical chemical defense, medical biological defense, clinical medicine, clinical surgery, physical protection, training, graduate medical education, and instruction.

Military activities that house, care, or use animals provided a work unit summary for any animal-based research. The FY97 BRD contained summaries and was made accessible to the public on October 1, 1998. A work unit summary may refer to a single protocol or a series of protocols that are performed in a given category of animal use. The summaries include the following information:

Title: Title of the work unit.

Funding Fiscal Year: The funding for the entire work for a given fiscal year. The funding includes civilian salaries, cost of animals, cost of materials, cost of human-based research, cost of non-animal based research, etc. — all costs related to the work unit except military salaries.

POC/Author: The primary contact (POC) for the work unit is usually the Public Affairs Office.

POC Address: The complete mailing address of the POC.

Performing Organization: The name of the activity where the work is performed.

Objective and Approach: This section is a narrative on the objectives and the approach of the work unit. This narrative provides a general summary of the work.

Indexing Terms (Descriptors): A list of indexing terms or keywords. The keywords contain “animals” and the term for any animal types that may be used in the work unit (e.g., guinea pigs, rats).

These summaries were compiled into the BRD and organized into a presentation format for the Internet.

II.3 ACCESS AND USE OF THE BIOMEDICAL RESEARCH DATABASE

The BRD can be accessed at:

<http://dticam.dtic.mil/dodbr/index.html>

The BRD home page shown in [Figure II-1](#) is a searchable database. To perform a search, click on Search. This will bring up the DoD BRD search page. The database can be searched by title,

keywords, description or specific demographic fields ([Figure II-2](#)). The results of the search will produce a hypertext list of titles ([Figure II-3](#)). To access a particular summary, click on the specific title and the summary will appear ([Figure II-4](#)).

II.4 FY98 UPDATE OF THE BIOMEDICAL RESEARCH DATABASE

The DoD will make all FY98 work unit summaries of animal use in research, testing, education, and training available to the public. All military activities that house, care, and/or use animals have provided summary information on any animal research, testing, education, or training work for the FY98 BRD. The cost of FY98 animal-based research is presented by work unit summary in the BRD. In order to prevent duplication, this information is not presented in this report. These data will become available to the public on October 1, 1999.

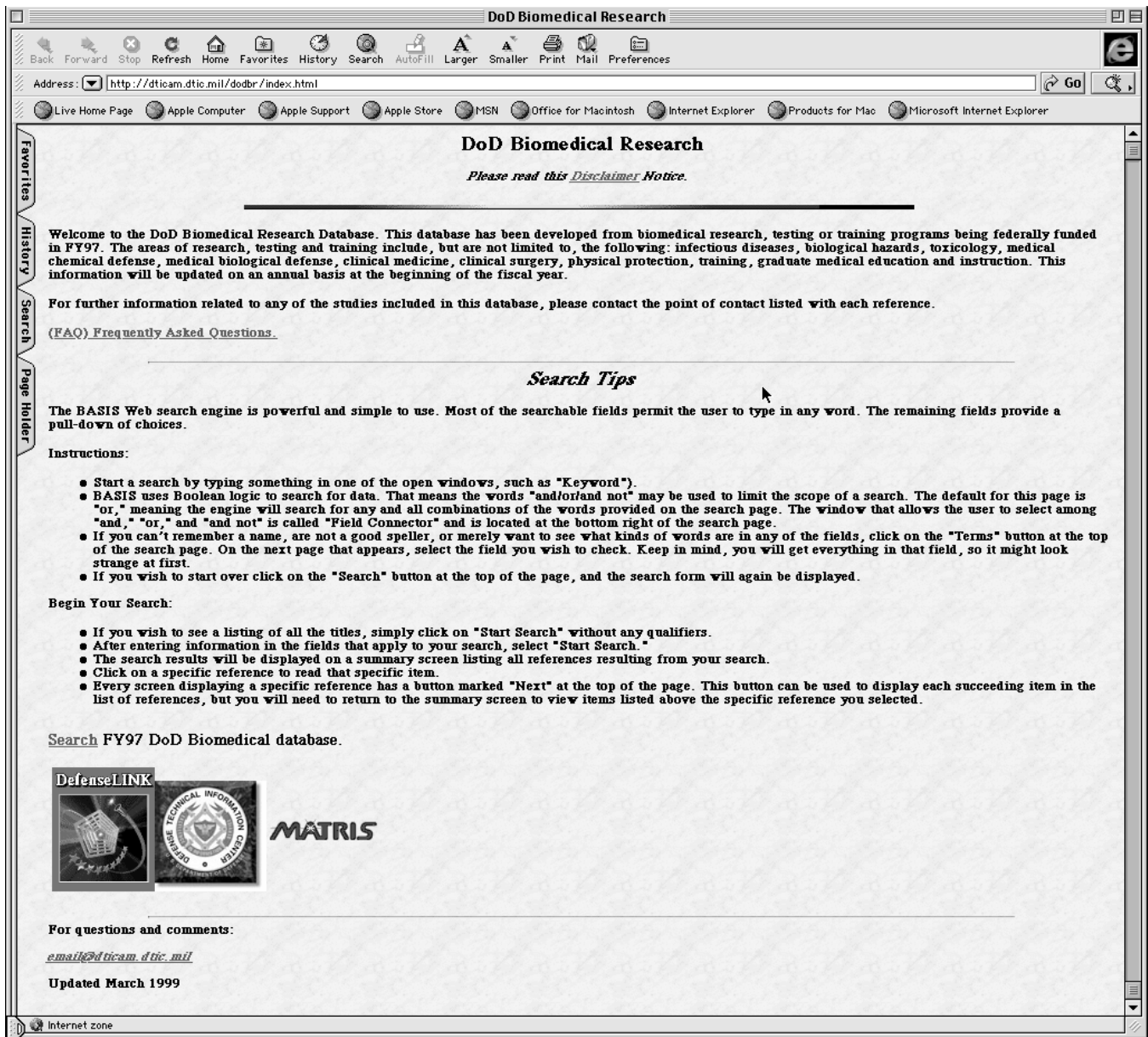


Figure II-1 DoD Biomedical Research Database Home Page

The screenshot shows a web browser window titled "<MATRIS.WWW>BIOWWW Search Form". The address bar displays "http://ocean.dtic.mil/BASIS/matrix/www/biowww/sf". The browser's menu bar includes Back, Forward, Stop, Refresh, Home, Favorites, History, Search, AutoFill, Larger, Smaller, Print, Mail, and Preferences. The toolbar contains icons for Live Home Page, Apple Computer, Apple Support, Apple Store, MSN, Office for Macintosh, Internet Explorer, Products for Mac, and Microsoft Internet Explorer. A vertical sidebar on the left contains links for Favorites, History, Search, and Page Holder. The main content area is titled "DoD BIOMEDICAL RESEARCH" and features a search form with the following elements:

- Buttons: "Start Search" and "Clear"
- Fields:
 - Keywords:** A text box containing the word "toxicity".
 - Title:** An empty text box.
 - Organization:** An empty text box.
 - Performing Organization:** An empty text box.
- Sorting options:
 - Order By:** A dropdown menu set to "NONE".
 - Sort Order:** A dropdown menu set to "Ascend".
 - Field Connector:** A dropdown menu set to "OR".
- Bottom navigation: A row of icons for BASIS, Search, Terms, Help, and Tips, followed by "Start Search" and "Clear" buttons.

The status bar at the bottom of the browser window indicates "Internet zone".

Figure II-2 DoD BRD Search Page

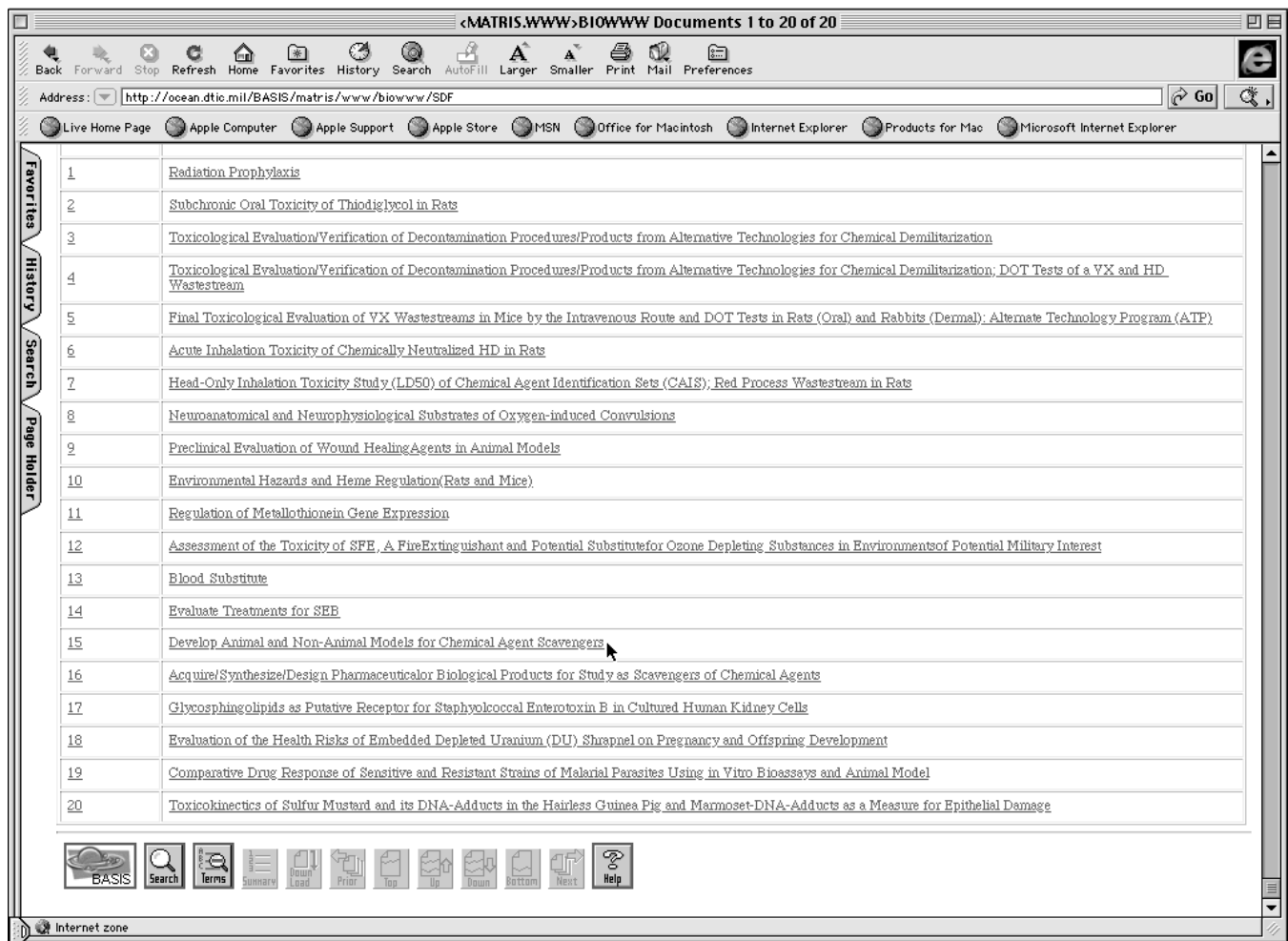


Figure II-3 Search Results on Toxicology from the BRD



SECTION III

OVERSIGHT OF DoD ANIMAL CARE AND USE PROGRAMS

This section of the Department of Defense (DoD) Report to Congress provides a detailed overview of the formal mechanisms and strategies for providing adequate oversight to the Department's numerous animal care and use programs. For the purposes of this report, research is defined as those congressionally authorized science and technology (S&T)-based activities—Title II, Research, Development, Test and Evaluation—of the Military Departments for which funds are appropriated within program elements 6.1 (Basic Research), 6.2 (Exploratory Development) and 6.3 (Advanced Development).

The mechanisms detailed here show a clear and long-standing commitment by the DoD to manage its animal-based research programs in a systematic, comprehensive, and effective manner. Individual programs are driven by specific mission requirements, and are subjected to a thorough, stratified review and analysis prior to commitment of funds. The DoD uses animals only when necessary to complete its mission, and in full compliance with applicable laws, regulations, and guidelines.

III.1 DETERMINATION OF DoD NEEDS FOR ANIMAL RESEARCH

Determining research needs and plans is a comprehensive process integrated into the DoD's planning, programming, and budgeting mechanisms. Integral elements of these processes are the Department's Research and Development Descriptive Summaries submitted to Congress in justification of the annual budget request. These summaries provide the Office of the Secretary of Defense, the Office of Management and Budget, and Congress with significant detail concerning the accomplishments and future plans of every research project.

Each DoD research laboratory employs its available resources to tailor its organization, staffing, and related infrastructure to best meet its S&T mission and to support the accountability, responsibility, and authority of its commander. In

October 1995, the Department implemented a comprehensive DoD Standard Protocol Format as a basis to justify and document all proposed animal use ([Appendix C](#)). The Standard Protocol Format solicits specific information that ensures a thorough review of all animal use proposals by Institutional Animal Care and Use Committees (IACUCs). Although there are minor differences in specific procedural elements in protocol review procedures among DoD facilities, DoD regulations ensure that the overall review mechanisms remain fundamentally similar. The general submission, review, and approval processes are summarized here.

An investigator develops a research protocol in support of Departmental S&T guidance and other supplementing instructions developed within the chain of command, both external and internal to the laboratory. Augmenting the formal S&T coordination and review process is a literature search to verify nonduplication of previous or ongoing research. The Standard Protocol Format requires that a search of Federal Research in Progress (FEDRIP), or its equivalent, and the Defense Technical Information Center (DTIC) database be made for DoD-funded research. An additional search of the scientific literature (MEDLINE, GRATEFUL MED, MEDLARS, AWIC, etc.) is highly recommended. Review and certification that this requirement has been met are integral elements of the review and approval process prior to initiation of a research project.

If animal use is planned for the intended research, the principal investigator must prepare an animal protocol request for submission to the facility IACUC. In addition to the DTIC and FEDRIP search, the Standard Protocol Format requires detailed information regarding results and dates of other on-line database searches (e.g., AWIC, AGRICOLA, CAAT, MEDLINE) that may yield alternatives to painful procedures. Additional pertinent knowledge and information on the proposed study are gained through review of the scientific literature and participation in scientific meetings, symposia, and workshops detailing other ongoing or completed research.

All individual protocols employing DoD resources are reviewed for factors such as military relevance, necessity, scientific merit, and relative research priority. These reviews are normally conducted within the laboratory's command-and-control structure and are characterized by the features of peer review systems.

DoD IACUCs carefully review research proposals involving the care and use of animals for numerous factors including, but not limited to ensuring that (a) the study is based on sound scientific principles; (b) a minimum number of animals are used to achieve the purpose; (c) the lowest phylogenetic species is selected as the appropriate model; (d) there is appropriate use of analgesics and anesthetics or, if required, there is adequate scientific justification for not using anesthetics; (e) the research is not duplicative; (f) the research personnel have the training and experience needed to conduct the research; and (g) the scientific question is of sufficient importance to warrant the use of animals. Additionally, detailed information regarding methodology, techniques, schedules, etc., is required, greatly facilitating a comprehensive and thorough review by IACUCs.

III.2 OVERSIGHT OF ANIMAL CARE AND USE PROGRAMS AND FACILITIES

There are three principal vehicles for oversight of animal care and use programs at DoD research facilities: Major DoD Activities and Service Command Staff, the local IACUC, and the Association for Assessment and Accreditation of Laboratory Animal Care (AAALAC) International.

III.2.1 Military Departments

Each military department has one or more components responsible for oversight and review of its research facilities and animal care and use programs. Periodic reviews, site visits, and inspections are conducted formally, and reports are prepared as required.

The Army's ultimate oversight responsibility is divided between two major commands: the U.S. Army Medical Command and the U.S. Army Materiel Command. In the U.S. Army Medical Research and Materiel Command, programmatic guidance and site visits are performed by veterinarians specialty trained laboratory animal medicine (LAM) in the Headquarters, U.S. Army

Medical Research and Materiel Command, and the U.S. Army Medical Department Center and School (Veterinary Programs Manager). In the U.S. Army Materiel Command, oversight is provided by a specialty trained LAM veterinarian assigned to the U.S. Army Chemical and Biological Defense Command.

Ultimate responsibility for laboratory animal care and use in the Navy is divided between the Office of the Chief of Naval Research and the Office of the Surgeon General of the Navy. Oversight for both offices is accomplished by a specialty trained LAM veterinarian assigned to the Naval Bureau of Medicine and Surgery. Besides biomedical research oversight for the Navy and the Marine Corps, this LAM veterinarian also serves the Naval School of Health Sciences, Bethesda (Clinical Investigations) and the Inspector General at the Naval Bureau of Medicine and Surgery.

U.S. Air Force responsibility for laboratory animal care and use is provided by the Office of The Surgeon General in addition to the Commanders of the Air Force Research Laboratory, medical centers, and the Air Force Academy. The U.S. Air Force Surgeon General's Research Oversight Committee (SGROC) monitors all animal use protocols, including both those performed at Air Force facilities and those contracted to civilian institutions. The SGROC approves all proposed research prior to initiation for projects involving nonhuman primates, companion animals, and marine mammals. A LAM veterinarian is assigned to the Air Force Surgeon General's Office to monitor the animal use research program and serves on the SGROC.

III.2.2 IACUCs

The backbone of the review process for all DoD animal-based research is the IACUC review of the research proposal or protocol. DoD Directive 3216.1, "The Use of Animals in DoD Programs," ([Appendix A](#)) requires all DoD facilities using animals in research to comply with the Animal Welfare Act (AWA). The AWA requires the Chief Executive Officer to appoint an IACUC, qualified through the experience and expertise of its members, to assess the research institution's animal program, facilities, and procedures. The AWA requires that IACUCs have a minimum of three members: an appropriately qualified chairman, at least one member not affiliated with the institution in any way other than as a member of the

Committee, and a veterinarian with training or experience in laboratory animal medicine and science. Each DoD IACUC is chaired by an individual with credentials and experience appropriate to the post, typically a senior physician, scientist, or veterinarian. DoD Directive 3216.1 clarifies the composition, membership, and training requirements of the IACUC. The 1995 revision to this Directive increased the minimum size of all DoD IACUCs from three to five, which is in concert with the National Institutes of Health (NIH) Office for Protection from Research Risks (OPRR) model. In addition, it specifies that

“...there shall be at least one non-scientific member on the IACUC. In addition, there shall be at least one member representing the general community interest who is nonaffiliated with the research facility. The nonaffiliated member and the non-scientific membership can be filled by the same person. To ensure community representation at each meeting and inspection, an alternate to the nonaffiliated member shall be designated for all IACUCs having a single nonaffiliated membership.”

The 36 IACUC panels reporting in FY98 averaged just over 8 members each. Private civilian, government civilian, and military representation on the panels is 9%, 44%, and 47%, respectively.

The diverse backgrounds/professions of the IACUC members are provided in [Figure III-1](#). Occupations/avocations for the nonaffiliated IACUC members are presented in [Appendix I](#). Currently, 31% of the nonaffiliated members are private sector civilians, the remainder are federal government civilians or military personnel. In accordance with DoD Directive 3216.1, these members represent the community and are not affiliated with (not under the command of) the research facility.

This Directive exceeds the requirements of the AWA and is further strengthened by the DoD 1995 Policy Memorandum ([Appendix B](#)) that directs a minimum of 8 hours of training for the new nonaffiliated members. DoD IACUCs implemented these requirements October 1, 1995. All DoD new nonaffiliated IACUC members received at least 8 hours of training to fulfill the requirement. The average total hours of training reported for

IACUC Membership by Occupation 306 Voting IACUC Members	
Professions	%
Research Scientists	29.4%
Veterinarians	23.5%
Other Nonscientists	19.0%
Physicians	13.4%
Animal Technicians	6.5%
Statisticians	3.3%
Other Medical*	2.6%
Ministry	2.2%

*Nurses, Dentists, Lab Technicians, Pharmacists

Percentages may not add up to 100% due to rounding of calculations.

Figure III-1 Background Professions of IACUC Members

nonaffiliated IACUC panel members increased from 11.8 in FY97 to 14.4 in FY98.

Each IACUC has at least one Doctor of Veterinary Medicine with training or experience in laboratory animal science and medicine who serves as an animal advocate. The U.S. Army Veterinary Corps' formal postgraduate training program in laboratory animal medicine provides didactic training in IACUC composition, function, and regulatory requirements. This training also prepares them to serve as animal advocates. The 37 DoD institutions reporting in FY98 reported an average of 2 to 3 veterinarians serving on their IACUC panels; 21 IACUC panels had 2 or more veterinarians.

It is a proactive Department policy that nonaffiliated members participate fully in discussions and vote on all research proposals. They are also encouraged to perform unannounced site visits of animal care facilities. In FY98, nonaffiliated members made 30 unannounced visits to Department animal facilities.

The IACUC has statutory responsibility for reviewing the facility's animal care and use program and inspecting the animal facilities on a semiannual basis. Consequently, at least once every 6 months, each IACUC performs an in-depth review

of the animal care and use program and inspects the animal facilities. To facilitate these inspections, the DoD has developed and implemented a standardized semiannual program review checklist that details the requirements of the review. All DoD IACUCs are currently using the new standardized checklist during their semiannual program reviews. The IACUCs prepare written reports of their evaluations and submit them to the Institutional Official, usually the facility commander. Reports specifically address compliance with the AWA, identify any departures from the Act, and include an explanation for the departure. The report must distinguish between major and minor deficiencies and provide a schedule for the resolution of deficiencies.

All DoD IACUCs document their meetings and activities, including the results of inspections, complaints, actions, and training. They are empowered to review and investigate concerns involving the care and use of animals at the research facility resulting from complaints received from the public or in-house workers, or from reports of noncompliance received from laboratory personnel. To facilitate the reporting and resolution of complaints or concerns, facilities commonly place signs or notices in high-traffic areas and in animal-study areas advising both the public and personnel who work with animals how to contact members of the IACUC, facility commanders, and/or the Inspector General (IG) whenever questions arise concerning humane care and treatment of animals. Among the reporting DoD institutions, three complaints were registered during FY98. DoD facilities have developed a wide variety of proactive and innovative mechanisms to inform the public how to contact responsible individuals and to ensure that those who work with animals are fully apprised of the requirement to provide humane and ethical care ([Appendix J](#)). Additionally, IACUCs make recommendations to the Institutional Official regarding any aspect of the research facility, its animal program, or the training of its personnel; review and approve, require modification to, or withhold approval of new research protocols involving the use of animals; review and approve, require modification to, or withhold approval of proposed significant changes regarding the care and use of animals in ongoing research protocols; and suspend an activity involving animals when they determine that the activity is not being conducted in accordance with its approved protocol.

III.2.3 AAALAC

AAALAC is a nonprofit organization chartered to promote high quality standards of animal care, use, and welfare through the accreditation process.

The AAALAC accreditation process provides scientists and administrators with an independent, rigorous assessment of the organization's animal care and use program. To increase accountability and tracking, a centralized DoD point of contact and database for AAALAC information have been established to enhance monitoring, reporting, and facilitation of the AAALAC accreditation process. An in-depth discussion of the AAALAC accreditation process and a profile of the DoD's participation are provided in [Section IV](#).

III.2.4 Training

The DoD provides extensive veterinary and animal care services for its facilities. Veterinarians with specialty training in LAM direct programs for animal care and use throughout the Department. They serve as a valuable resource to the research staff and the IACUC to ensure that all research methods and maintenance procedures are consistent with the latest principles of animal medicine, and with the current interpretations and implementing regulations of the AWA. The DoD sponsors formal postdoctoral training programs for veterinarians in LAM, including a nationally recognized, in-house 2-year residency program culminating in specialty board eligibility for certification by the American College of Laboratory Animal Medicine. Many DoD veterinarians attend various university postgraduate LAM training programs resulting in a master's degree in public health or Ph.D. It is significant that approximately 25% of the current membership of American College of Laboratory Animal Medicine, the veterinary specialty most closely associated with animal welfare and laboratory animal care and use, received either all or part of their training in DoD-sponsored LAM training programs. In August 1995, the DoD began a formal postgraduate Master's of Public Health in Laboratory Animal Medicine at the Uniformed Services University of the Health Sciences. This outstanding program provides the Department with a new source of LAM experts who will significantly enhance animal welfare in our research laboratories.

In addition to veterinarians, the DoD trains animal care specialists (Military Occupation Specialty 91T) to assist in the daily management, care, and treatment of laboratory animals. Over the last 31 years, the DoD has trained over 3,600 animal care specialists. Since 1986, the Division of Veterinary Medicine has sponsored the Walter Reed Army Institute of Research (WRAIR) DoD Laboratory Animal Workshop program. Many of the workshops focus on species-specific techniques and handling, while others provide general laboratory animal information required by federal law and other guidelines for the research mission. Successful completion of the workshops fulfills the training requirements for use of those animals in research protocols. The WRAIR DoD Laboratory Animal Workshop trained over 400 investigators and technicians in FY98, and the course schedule is provided in [Appendix K](#). Additionally, DoD research institutions send appropriate staff to a variety of seminars and workshops sponsored by the National Institutes of Health, other federal agencies, and private institutions dedicated to the proper care and use of research animals. The Annual Public Responsibility in Medicine and Research Meeting is an outstanding example of this type of training.

The DoD provides detailed informational and instructional material to all members of the IACUC, including nonaffiliated members, to ensure that they are fully cognizant of the numerous responsibilities of IACUC members under the provisions of the AWA. DoD Directive 3216.1 “The Use of Animals in DoD Programs” requires new nonaffiliated IACUC members to receive an initial 8 hours of training and continued training for IACUC members, investigators, and technicians. This requirement went into effect October 1, 1995. Although training is an individual institute’s responsibility, the DoD has developed a program consisting of a set of topics and recommended resources to support the training requirement ([Appendix L](#)). The topics are meant to be general and allow for tailoring of the training to meet the institute’s specific needs. The recommended resources are readily available commercially. Formal training on animal care and use issues is provided to all appropriate personnel in Department research laboratories in accordance with the provisions of the AWA. Examples of training or materials currently provided to IACUC members are detailed in [Appendix L](#). One of the examples listed in [Appendix L](#) is the Institute

of Laboratory Animal Research (ILAR) publication *Education and Training in the Care and Use of Laboratory Animals*. As one of the major sponsors of this publication, the DoD has established a formal relationship with the National Research Council (NRC), an extension of the National Academy of Sciences. The publication is used as a guide by the DoD and has been translated into five languages. Many countries use this publication as a standard for the care and use of laboratory animals.

III.2.5 Community Visits

Individuals or groups wishing to visit Department facilities need to comply with certain procedural guidelines. All DoD facilities are served by a public affairs office, at either the facility, post, or base. Visits by the public or the press are arranged and coordinated through the appropriate public affairs office. While most facilities reported few community visits, 48 community visits were described in FY98. DoD facilities are visited by various special interest groups including community and civic groups; animal welfare or animal advocates, groups, or individuals; dignitaries, academia, and teachers; local, state, and national politicians; congressional members and staff; elementary to postdoctoral students; etc. Consequently, a greatly diversified range of individuals is constantly visiting and observing the quality of Department facilities.

III.2.6 Office for Protection from Research Risk Oversight

A number of DoD research laboratories participate in the NIH grants process. Institutional compliance with The Public Health Service Policy on Humane Care and Use of Laboratory Animals (PHS Policy) is a prerequisite for granting or continuation of NIH intramural and extramural funding. The formal vehicle for compliance with the PHS Policy is an “Animal Welfare Assurance” negotiated between individual institutions and the OPRR. The principal references for the negotiation of an OPRR “assurance” are the Health Research Extension Act of 1985 (Public Law 99-158, November 20, 1985, “Animals in Research”), the Animal Welfare Act, and NRC’s *Guide for the Care and Use of Laboratory Animals*. Consequently, OPRR provides additional oversight to those laboratories that have negotiated OPRR assurances.

III.2.7 Additional Oversight

Within the DoD, individuals may raise animal welfare concerns. This may be with the IACUC, facility commanders, the IG, or the attending veterinarian. Other means of compliance or concern may be voiced through “Waste, Fraud and Abuse Hotlines,” or the formal chain of command. Procedures to enhance and facilitate these mechanisms have been implemented in DoD facilities.

The function of the IACUC and the role of an ombudsman are augmented by the Department’s IG. An ombudsman is defined in Webster’s dictionary as “a government official charged with investigating citizens’ complaints against the government.” The Humane Society of the United States, a witness at the April 7, 1992 hearing on The Use of Animals in Research by the Department of Defense before the House Armed Services Committee, offered the Ombudsman Program at the Massachusetts Institute of Technology as an example of a model program. This program consists of an ombudsman assigned to the university president’s office to hear complaints regardless of the nature. These include personnel complaints, sexual harassment, animal welfare, etc. The DoD assigns this responsibility to its IG and respective Inspectors General (IGs) of the Military Departments. In addition, military bases and large organizations on military bases have their own IGs who fulfill this function. Significantly, complaints to an IG can be made anonymously. Also of note is the fact that IG investigations are conducted with complete autonomy, and are completely insulated and immune to pressure from the chain of command.

Oversight of extramural (contract) animal-based research is provided for in DoD Directive 3216.1 (1995) ([Appendix A](#)). It states that

“a. all extramural research proposals using live animals shall be administratively reviewed by a DoD veterinarian trained or experienced in laboratory animal science and medicine before grant or contract award.

“b. the most recent [U.S. Department of Agriculture] USDA inspection reports are provided or obtained for the facility under consideration for a research contract or grant using animals, and that during the term of the award, the most recent USDA inspection reports be reviewed on an annual basis.

“c. a DoD veterinarian trained or experienced in laboratory animal science and medicine shall conduct an initial site visit to evaluate animal care and use programs at contracted facilities conducting DoD-sponsored research using nonhuman primates, marine mammals, dogs, cats, or proposals deemed to warrant review. The initial site visit shall occur within 6 months of when the facility has taken delivery of the animals under DoD contract or grant award. Any facility receiving a DoD-funded grant or contract for animal-based research shall notify the DoD component sponsor and shall have a site inspection within 30 days of notification of loss of AAALAC accreditation for cause, or notification that the facility is under USDA investigation. Site inspections for cause shall evaluate and ensure the adequacy of animal care and use in DoD-sponsored programs, and provide recommendations to the sponsoring DoD component about continued funding support of the research.”

As directed by DoD Directive 3216.1, all nonhuman primate protocols receive an additional centralized review external to the research facility.

III.3 CHAIN OF COMMAND OVER ANIMAL CARE AND USE PROGRAMS

The chain of command is designed to resolve problems at the lowest possible level. It provides control and communication among various components of organizations. Each link in the chain of command is a level of responsibility and authority that extends from the President of the United States, as Commander in Chief, down to the lowest supervisory level. Different levels within the chain have different responsibilities and authority. Each level in the chain is responsible for a lower level and accountable to a higher one. Every individual in the military is part of the chain of command and is accountable to it.

III.4 AVOIDANCE OF UNINTENDED DUPLICATION OF RESEARCH

Both the DoD and Congress have a long history of concern about the potential for unintended duplication of Defense research. Within the past decade, the Department has initiated significant improvements in its mechanisms for coordination, and joint planning and review of its research programs.

In 1981, Congress expressed concerns about the potential for unnecessary duplication of biomedical research among the Military Departments (H.R. 96-1317). This resulted in the DoD proposing an Armed Services Biomedical Research Evaluation and Management (ASBREM) Committee to coordinate biomedical research planning and the conduct of biomedical research among the Military Departments. Congress fully endorsed and built upon this proposal by establishing DoD Lead Agencies for major elements of the biomedical research programs for which there were either no, or very few, service-unique requirements (H.R. 97-332). For example, the Army was designated the DoD Lead Agency for military infectious disease and combat maxillofacial research while the Navy was designated DoD Lead Agency for preventive and emergency dentistry research. The ASBREM Committee established Joint Technology Coordinating Groups (JTCGs) (Figure III-2), consisting of directors of biomedical research programs and representatives of biomedical research laboratories, to coordinate all DoD biomedical research planning and execution. The

ASBREM Committee process has proven to be highly effective at eliminating unnecessary duplication of biomedical research.

Because of the wide range of organizations and variations in process between the Military Departments and Defense Components, the DoD uses a variety of mechanisms to coordinate its research and training. The ASBREM Committee process became the model for joint DoD coordination initiatives. Responsibility for joint coordination, planning, execution, and review of the Department's S&T programs was assigned to joint oversight bodies: the Joint Directors of Laboratories (JDL), the ASBREM Committee, the Training and Personnel Systems Science and Technology Evaluation and Management (TAPSTEM) Committee, and the Joint Engineers. The resulting technology area responsibilities are shown in Figure III-3. The JDL is responsible for general oversight as well as specific joint planning for Combat Materiel (Figure III-4). TAPSTEM oversees DoD personnel and training research (Figure III-5) and the Joint Engineers oversee environmental quality and civil engineering

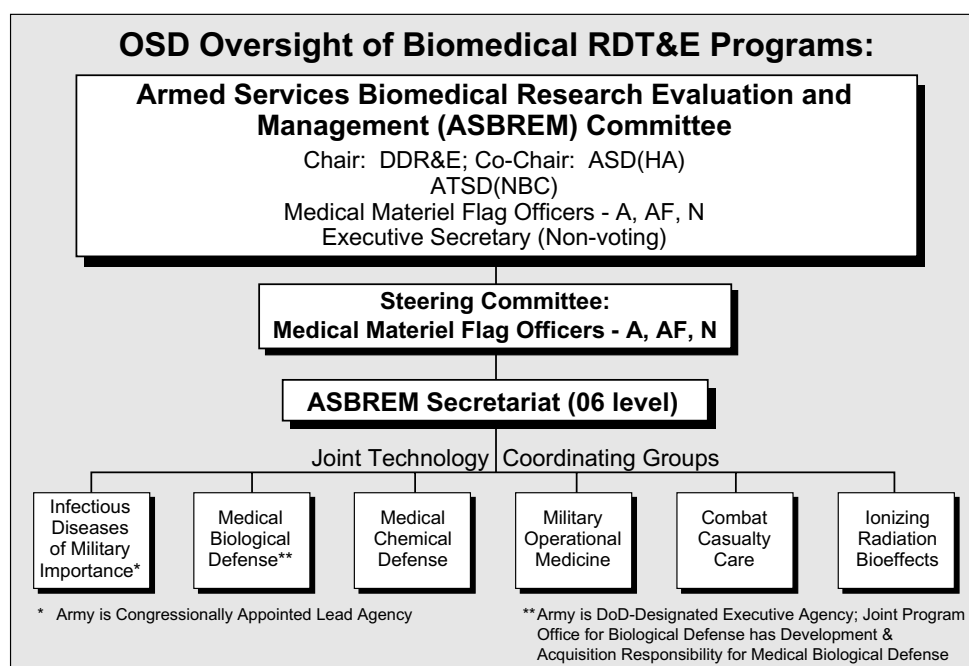


Figure III-2 Structure of ASBREM Committee

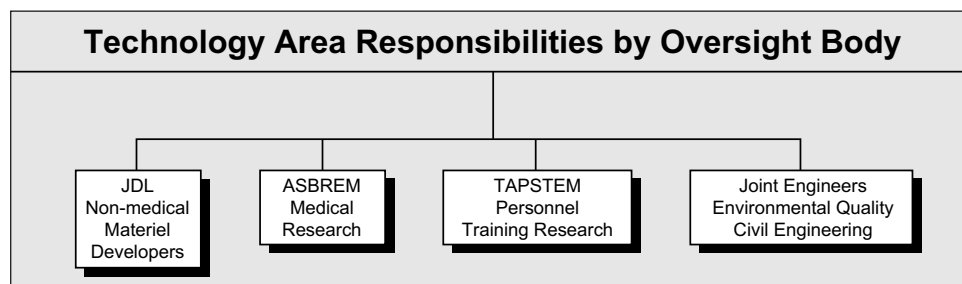


Figure III-3 DoD Technology Area Responsibilities

(Figure III-6). These oversight bodies are assisted in execution of their responsibilities by subordinate S&T coordinating groups that are focused on coordination of specific technology areas. For example, the ASBREM and TAPSTEM Committees are supported by the JTTCGs, (Figures III-2 & 5) and the JDL and Joint Engineers are supported by separate technology panels (Figures III-4 & 6). Under this process, researchers and managers from the service laboratories jointly plan execution and coordinate their research to minimize redundancy and take advantage of each others strengths.

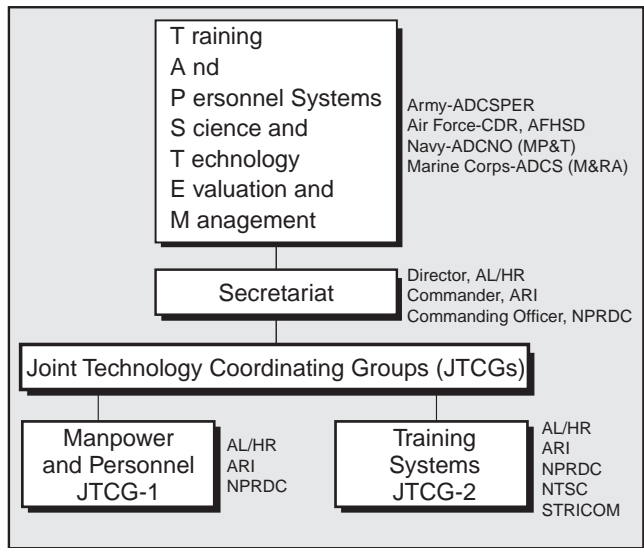


Figure III-4 TAPSTEM Organization

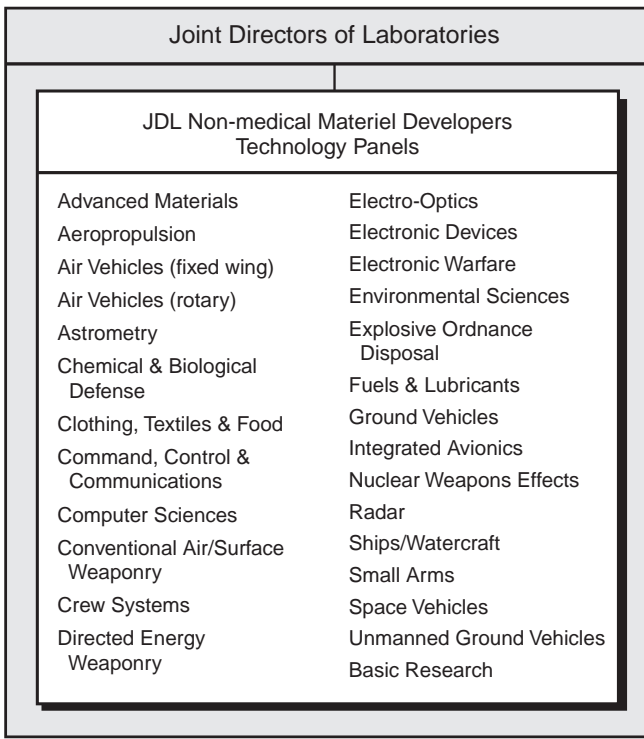


Figure III-5 JDL Technology Panels

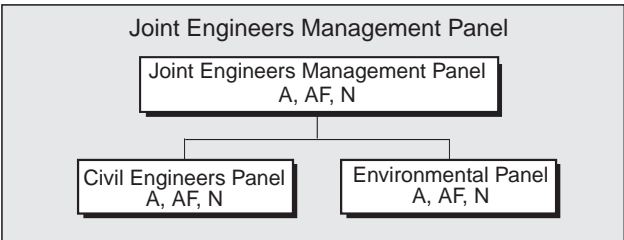


Figure III-6 Joint Engineers Management Panel

In addition to these formal coordination and review processes to eliminate research duplication, there are a number of less formal mechanisms that provide significant disincentives for research duplication. Competition, both in-house and extramural, for research support is a prominent feature of S&T; each year large numbers of scientifically meritorious research proposals cannot be funded due to shrinking resources and funding shortages. In most cases, the professional stature of individual scientists or engineers among their peers is measured by their individual and original contributions to the scientific literature. There is little if any reward for unnecessarily duplicating the work of others; such actions often have significant negative impact on how the scientist or engineer is viewed by peers and on the ability to secure research support. Additionally, within the DoD civilian personnel system, scientists' and engineers' pay grades are determined in part by the level of individual scientific and technological contributions. One outcome of research is publication of a manuscript in a professional journal. A sample listing of journals with DoD animal research publications is found in Appendix G. Peer-reviewed journals critique the research during the review process, leading to an overall enhancement of the research process and to validation of both the scientific merit and necessity of the research. These less formal, relatively unquantifiable, disincentives substantially augment and buttress the Department's formal mechanisms for regulating and avoiding unnecessary research duplication within its S&T programs.

III.5 AVOIDANCE OF UNNECESSARY RESEARCH

The same factors that effectively prevent unwarranted duplication of research are applied to prevent unnecessary research. Additionally, through Cooperative Research and Development Agreements, the Department has increased its emphasis on leveraging and exploiting, for Defense

needs, S&T investments from other federal agencies, U.S. industry, and academic institutions, and the international scientific community. Past descriptions of Defense S&T “spin-off” have been supplanted by programs intended to “spin-on” accomplishments by others as well as to optimize the dual-use potential of the Defense S&T investment. The foundation of Defense S&T strategy is the application of S&T accomplishments to sustain Defense technological superiority through efficient and responsive modernization of our warfighting capabilities.

III.6 SUMMARY

Research using animals is highly structured and regulated in the United States, being governed by numerous laws, regulations, and policies. Consequently, the DoD has a number of stratified formal and informal mechanisms for reviewing, regulating, and executing its animal care and use programs. Research performed by the DoD receives close programmatic, scientific, and regulatory scrutiny, being carefully reviewed by various offices, committees, and program managers before it is funded or implemented. These reviews serve to determine the necessity to the mission, provide

oversight of animal care and use, and avoid unnecessary or unintended duplication of research.

Individual IACUCs provide oversight of animal care and use programs and research. They also provide training and information about animal care and use, and ensure the humane use of animals in research. Each DoD facility’s IG is also an effective means for investigation of concerns about the necessity of animal use, as well as the ethical treatment and humane care of animals used in DoD research.

Over the past decade, the DoD, in concert with Congress, has streamlined and greatly improved coordination of its S&T activities to avoid unnecessary duplication and provide a focused program of research responsive to the DoD’s unique and wide-ranging needs.

When viewed in its totality, the Department’s significant progress and investment in administration, infrastructure, standardization, training, and oversight of animal use are indeed impressive, and can serve as useful models for the rest of the animal use research community.

SECTION IV

AAALAC ACCREDITATION OF DoD LABORATORIES

The Department of Defense (DoD) recognizes the benefits of accreditation by the Association for Assessment and Accreditation of Laboratory Animal Care, International (AAALAC). With the publication of the Joint Regulation on the Use of Animals in DoD programs, June 1, 1984 (AR 70-18), the DoD implemented more stringent animal care and use requirements than those required by statute. The Joint Regulation established uniform procedures, policies, and responsibilities for the use of animals in the DoD. The DoD has elevated the requirement with the current DoD Directive 3216.1 (1995), which states that “all DoD laboratories that maintain animals for use in research, testing or training shall apply for AAALAC accreditation.” The Joint Service Regulation also cites the National Research Council (NRC) publication, *Guide for the Care and Use of Laboratory Animals*, which is the principal document used by AAALAC in its accreditation process. The animal care and husbandry standards and requirements contained in the Guide are designed to provide an environment that ensures proper care and humane treatment are given to all animals used in research, testing, and training. This care requires scientific and professional judgment based on knowledge of the husbandry needs of each species, as well as the special requirements of the research program.

IV.1 AAALAC ACCREDITATION

AAALAC accreditation is widely accepted by the scientific community, and viewed as an extremely desirable feature of the Department’s animal care and use programs. The Association is highly respected as an independent organization that evaluates the quality of laboratory animal care and use. Accreditation covers all aspects of animal care to include institutional policies; laboratory animal husbandry; veterinary care; facility physical plant; support facilities; and special areas of breeding colony operations and animal research

involving hazardous agents such as radioactive substances, infectious agents, or toxic chemicals.

The independent and external peer review that is fundamental to continuing AAALAC accreditation is valuable to any program. All AAALAC findings highlight program strengths and identify potential weaknesses. Laboratories maintaining accreditation demonstrate a high degree of accountability and program excellence. AAALAC standards stress the appropriate appointment, composition, and empowerment of an Institutional Animal Care and Use Committee (IACUC). This Committee is responsible for monitoring and evaluating all aspects of the institution’s program that use animals for teaching and/or research purposes. IACUC functions are addressed in [Section III](#) of this report.

IV.2 DoD PROGRAM REVIEWS

The DoD utilizes external peer review by the Joint Commission for Accreditation of Health Organizations to evaluate many of its programs such as drug screening laboratories and military medical facilities. At the same time, the DoD recognizes the diversity of mission operations and the global reach of the military mission. There are situations where external peer reviews are not cost effective due to remote locale, limited scope of operations, or host nation sovereignty. In these cases, equivalency standards can be applied and effectively monitored. The Joint Service Regulation and Service-conducted inspections of facilities implement the requirements of the Animal Welfare Act and the 1996 NRC *Guide for the Care and Use of Laboratory Animals*.

The DoD is committed to continuing its full participation in the AAALAC accreditation process in order to effect external peer review for assessing program compliance with regulations, guidance, and ethical responsibility.

IV.3 DoD AAALAC ACCREDITED PROGRAMS

The number of DoD AAALAC accredited programs that maintain animals for research testing and training has significantly increased over the past 6 years (Figure IV-1). Of the 36 DoD facilities

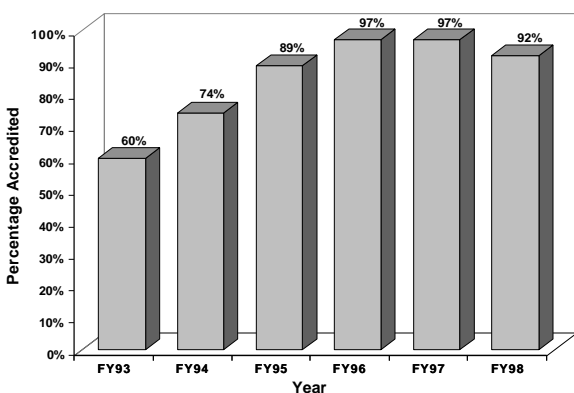


Figure IV-1 DoD AAALAC Accreditation at Time of Publication of the FY93-98 Reports

worldwide reporting animal use, 33 (92%) are AAALAC accredited. In FY98 there were two new facilities that used animals (Landstuhl Regional Medical Center in Germany and Camp Lejeune in North Carolina). Both of these facilities were not AAALAC accredited. The percentage decrease from FY97 (97%) to FY98 (92%) is directly related to these new facilities using animals in FY98.

The three programs not accredited in FY98 are Landstuhl Regional Medical Center in Germany, the Armed Forces Research Institute of Medical Sciences in Thailand, and Camp Lejeune in North Carolina. The first houses animals only occasionally and transiently (less than 24 hrs.), the second is undergoing completion of a 5 million dollar laboratory animal facility renovation, and the third has since eliminated all animal use in favor of employing artificial models for medical training.

These efforts reflect the DoD's commitment to accrediting all of its animal care and use programs.

IV.4 AAALAC ACCREDITATION STATUS FOR U.S. DoD PROGRAMS

There are 31 programs in the United States that maintained animals for research, testing, or training for the DoD in FY98. With the exception of Camp Lejeune (which has eliminated animal use programs in the future), in FY98 all programs in the U.S. were accredited by AAALAC. In addition, there are four DoD animal use programs that share DoD AAALAC accredited facilities. These programs are small detachments that are assigned to DoD bases and therefore share their animal care and use facilities. [Appendix M](#) provides additional information on AAALAC accreditation by program.

IV.5 AAALAC ACCREDITATION STATUS FOR DoD OVERSEAS PROGRAMS

There are five DoD programs using animals outside the United States. In foreign countries, the accreditation process is often complicated by issues of sovereignty; local governments have their own regulations and policies that must be considered. Renegotiation of various agreements may be involved in construction or renovation projects. Despite these and various other impediments, the DoD has raised the standard of excellence in its animal care and use programs by receiving full accreditation in four of its five overseas laboratories. The Naval Medical Research Detachment in Lima, Peru, was the first laboratory in South America to have received AAALAC accreditation. The Naval Medical Research Unit #2 in Jakarta, Indonesia, and the Naval Medical Research Unit #3 in Cairo, Egypt were the first to be accredited in Southeast Asia and Africa, respectively. Facilities of the Armed Forces Research Institute of Medical Sciences are anticipated to receive accreditation in a scheduled AAALAC site visit in 1999.

SECTION V

DoD ANIMAL USE PROFILES

The information presented in this section provides profiles on the reported use of animals in various research categories, and the U.S. Department of Agriculture (USDA) pain categories of Department of Defense (DoD) animal-based research, testing and training programs for fiscal year (FY) 1998.

V.1 METHODS

Information was solicited and received from DoD agencies and military commands, organizations, and activities involved in animal care and use programs located both inside and outside of the United States. These included extramural contractors and grantees that performed animal-based research. For the purpose of this reporting requirement, an intramural program represents research performed at a DoD facility and funded by either DoD or non-DoD funds. An extramural program represents research performed by a contractor or grantee that is funded by the DoD.

V.1.1 Animal Use Profiles

The animal use profiles prepared for this report are consistent with the reporting information and data provided to the USDA using Animal and Plant Health Inspection Service (APHIS) Form 7023. In addition, this report contains comprehensive information on all other animals (e.g., mice, rats, birds) used that are not required in reports to the USDA.

For the purposes of this reporting requirement, an animal was defined as any whole nonhuman vertebrate, living or dead, excluding embryos, that was used for research, development, test, and evaluation (RDT&E), clinical investigations, diagnostic procedures, and/or instructional programs. Only live animals or whole dead animals, as defined, that were either on hand in the facility or acquired during FY98 and used are included. Animal organs, tissues, cells, blood, fluid components, and/or byproducts purchased or acquired as such animal/biological components are

not reported. This definition does not include animals used or intended for use as food for consumption by humans or animals, animals used for ceremonial purposes, or military working animals and their training programs.

A single animal was counted only once in determining the number of animals used during the fiscal year for a particular work unit or protocol. This does not refer to the number of times an individual animal was injected, manipulated, handled, or administered medication and/or experimental compounds within a given work unit, protocol, or program. Animals on hand during FY98, but not actually used during the fiscal year, are not included in this number.

V.1.2 Animal Use Categories

All DoD agencies and military commands, organizations, and activities involved in the performance and/or funding of animal care and use programs reported animal work by the category that best describes the general research purpose of the animal use. The 8 general categories and 23 specific subcategories are listed in [Table V-1](#). If the research categories provided did not adequately describe the animal use within each particular work effort, the animal was placed in the Other category. In-depth information on specific activities performed within a subcategory is presented in [Appendix N](#). The medical research categories correspond to the Armed Services Biomedical Research Evaluation and Management (ASBREM) Committee's Joint Technology Coordinating Group Medical Research Areas. Non-medical categories consist of RDT&E programs performed outside the ASBREM Committee medical oversight. Clinical Investigations studies were performed under the auspices of the Assistant Secretary of Defense for Health Affairs and the military services medical departments through Major Force Program 8 funding. These studies were usually in support of graduate medical education training programs located at the major military medical centers.

Table V-1 Animal Use Categories

MEDICAL (M)
M1: Military Dentistry
M2: Infectious Diseases
M3: Medical Chemical Defense
M4: Medical Biological Defense
M5: Human Systems Technology
M6: Combat Casualty Care
M7: Ionizing Radiation
M8: Other Medical RDT&E
NON-MEDICAL (N)
N1: Physical Protection
N2: Physical Detection
N3: Offensive Weapons Testing
N4: Other Non-Medical RDT&E
CLINICAL INVESTIGATIONS (C)
C1: Clinical Medicine
C2: Clinical Surgery
C3: Other Clinical Investigations
TRAINING/INSTRUCTIONAL (T)
T1: Training, Education, and/or Instruction of Personnel
T2: Other Training/Instruction
ADJUNCTS/ALTERNATIVES TO ANIMAL STUDIES (A)
A1: Adjuncts to Animal Use Research
A2: Alternatives to Animal Investigation
A3: Other Alternatives/Adjuncts
CLASSIFIED SECRET OR ABOVE STUDIES (S):
Classified secret or above studies on animals
ANIMAL BREEDING STOCK (B): Animals maintained for breeding
OTHER ANIMAL USE CATEGORIES (O): Other animal use purposes

Table V-2 USDA Pain Categories
(USDA APHIS Form 7023)

USDA COLUMN C
Number of animals upon which teaching, research, experiments, or tests were conducted involving no pain, distress, or use of pain-relieving drugs.
USDA COLUMN D
Number of animals upon which experiments, teaching, research, surgery, or tests were conducted involving accompanying pain or distress to the animals and for which appropriate anesthetic, analgesic, or tranquilizing drugs were used.
USDA COLUMN E
Number of animals upon which teaching, experiments, research, surgery, or tests were conducted involving accompanying pain or distress to the animals and for which the use of appropriate anesthetic, analgesic, or tranquilizing drugs would have adversely affected the procedures, results, or interpretation of the teaching, research, experiments, surgery, or tests.

without anesthesia or analgesia. Examples include most blood-sampling techniques (excluding intracardiac and periorbital blood sampling), injections, and tattooing.

The animals reported in Column D of the USDA report are those that experience pain in which appropriate anesthetic, analgesic, or tranquilizing drugs were used. Examples include anesthesia for surgical procedures or catheter placement, and analgesia during recovery from surgery.

The animals reported in Column E of the USDA report are those that would experience more than slight or momentary pain or distress that cannot be alleviated by drugs. Examples of procedures where drugs were not used because they would have adversely affected the procedures, results or interpretation of the research, or tests include some infectious disease studies and some toxicology studies.

All procedures that involve animals in USDA Pain Category Columns D or E are extensively reviewed during the protocol approval process. Prior to formal protocol review, a veterinarian with experience and/or training in laboratory animal medicine must review all procedures that could cause pain and distress in animals. In addition, the primary investigator must write a justification for all procedures for animals in Columns D and E. The DoD standard protocol states, "Procedures causing more than transient or slight pain that are

V.1.3 USDA Pain Categories

The USDA requires that all institutions using any regulated animal for research, testing, training, or experimentation register with the USDA as a research facility and submit an annual report. This annual report presents the number of regulated animals used and the type of pain, if any, to which the animals were exposed.

The USDA has developed three pain categories for its reporting requirement (Table V-2). All animals herein reported are assigned to one of the three USDA pain categories; this includes animals that are not regulated by the USDA. The USDA requires that any reporting facility that uses procedures producing unalleviated pain or distress file an explanation of the procedures with its annual APHIS report.

The animals reported in Column C of the USDA report are those used in procedures that are not painful. Procedures performed on these animals are those that are usually conducted on humans

unalleviated must be justified on a scientific basis in writing by the primary investigator. The pain must continue for only the necessary period of time dictated by the experiment, and then be alleviated, or the animal humanely euthanized.” Moreover, the primary investigator must sign an assurance statement that alternative procedures are not available, and the Institutional Animal Care and Use Committee must review and approve all procedures before the study begins.

V.2 RESULTS/DISCUSSION

V.2.1 General Results

There was a total of 291,551 animals reported used in FY98, which is an 8% decrease from FY97 and a 47% decrease from FY93 (Figure V-1). The Animal Welfare Act of 1985 defines animals as “any live or dead dog, cat, monkey (nonhuman primate mammal), guinea pig, hamster, rabbit, or such other warm-blooded animal, as the Secretary may determine.” Therefore, only 9% (26,750) of the animals reported used by the DoD in FY98 are considered USDA reportable species.

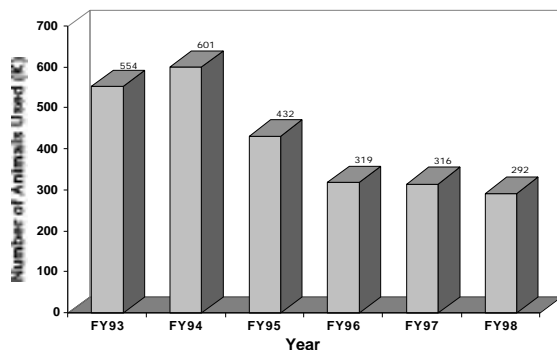


Figure V-1 DoD Animal Use by Year

In FY98, 152,824 animals were reported used in intramural research programs and 138,727 were used in extramural grants or contracts (Figure V-2). Reported intramural animal use increased by 2%, (3,637) in FY98 compared with FY97 use and decreased by 43% (115,267) compared with FY94 use. The number of animals reported used in extramural research was 17% (28,134) lower in FY98 than the number in FY97 and 58% (193,865) less than the number used in FY94. Extramural programs by their very nature have large fluctuations in the number of animals used from year to year. Each year a different number of contracts are granted to perform extramural research. Many of these do not use animals at all; others only use animals during a

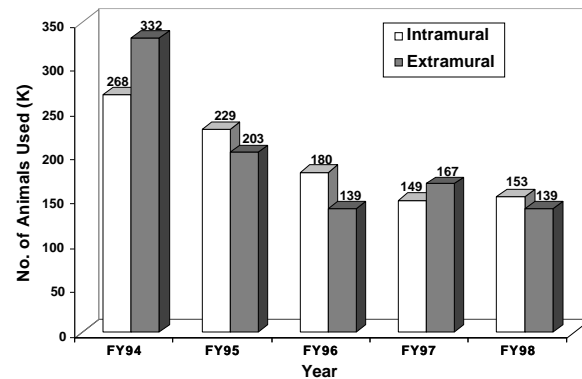


Figure V-2 Intramural/Extramural Animal Use by Year

portion of the proposed project (e.g., third year of project); and others use animals throughout the entire project. In addition, the level of funding for extramural programs varies from year to year, thereby changing the total number of extramural projects. Some extramural research programs are congressionally mandated such as Breast Cancer, Gulf War Illnesses, Neurofibromatosis, and Osteoporosis Research Programs; their funding is dependent on yearly congressional appropriations. Therefore, changes in the number of animals used by the DoD extramural research programs can fluctuate significantly from year to year. The intramural programs have less variation in their use of animals because they have a continuous mission and ongoing research in specific areas. Consequently, any decrease in the number of animals used is most likely a result of the use of alternatives to animal use, a decrease in the number of research projects, or a decrease in intramural funding.

V.2.2 Animal Use by Service

Information concerning total reported DoD use of animals by each service is presented in Figure V-3. Figures V-4 and V-5 show the intramural and extramural animal use by service, respectively.

In FY98, the Army used 72% of the total number of animals reported used by the DoD, 59% of the intramural animals, and 85% of extramural animals. There was a 6% decrease in the Army's reported intramural animal use and a 9% decrease in extramural animal use since FY97. The Army has an ongoing responsibility to manage the congressionally mandated Breast Cancer, Prostate Cancer, Neurofibromatosis, Osteoporosis, and Defense Women's Health Research Programs. These programs used

TOTAL = 291,551

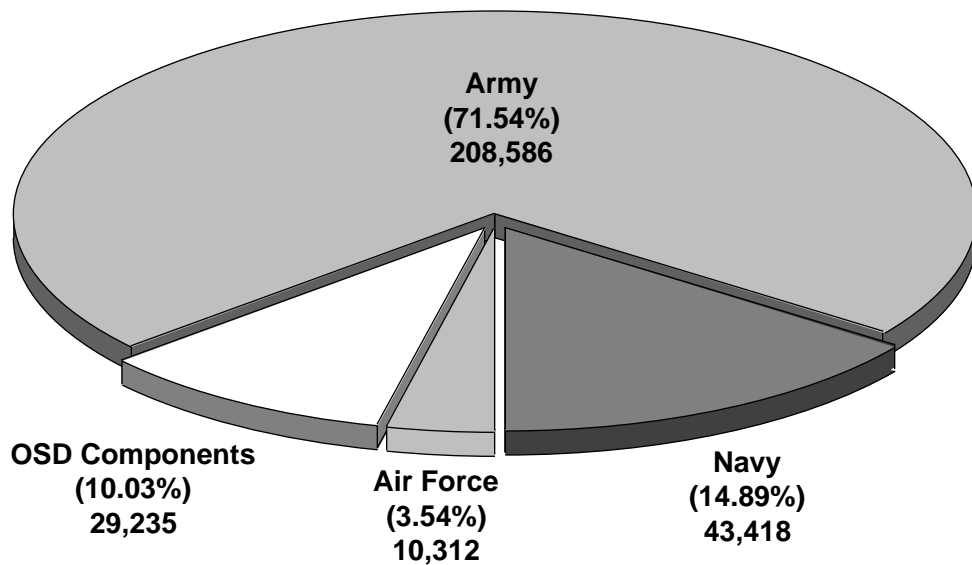


Figure V-3 DoD Intramural and Extramural Animal Use by Service for FY98

TOTAL = 152,824

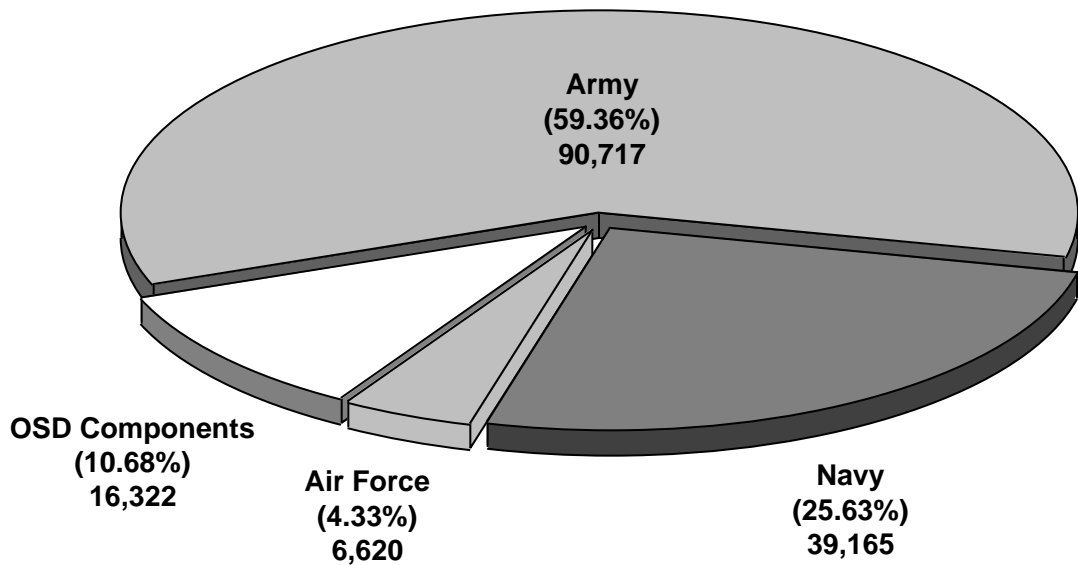


Figure V-4 DoD Intramural Animal Use by Service for FY98

Percentages may not add up to 100% due to rounding of calculations

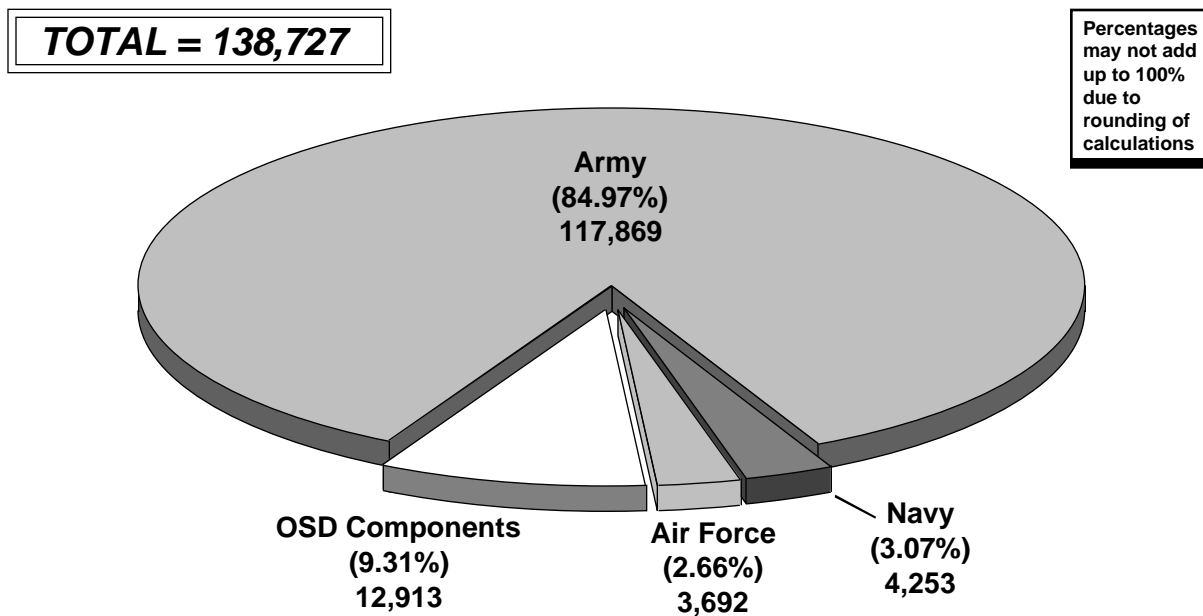


Figure V-5 DoD Extramural Animal Use by Service for FY98

the majority of the Army's extramural research animals (54,283). In addition, 1,167 animals were used in research on Gulf War Illnesses. The U.S. Army Medical Research and Materiel Command is the congressionally mandated Lead Agency for infectious disease and combat dentistry research and the DoD Executive Agency for medical chemical and biological defense and nutrition studies. The number of animals the Army used in research on infectious diseases and chemical and biological defense was 31,957 and 84,522, respectively. Overall, the Army had an 8% decrease in animal use between FY97 and FY98.

The Navy used 15% of the total number of animals reported used by the DoD, 26% of the intramural animals, and 3% of extramural animals. Comparing reported animal use in FY98 with use in FY97, there was a 65% (17,066) increase in the total number of animals used by the Navy. This increase was in the Navy's intramural research projects, which increased by 98% (19,357). The Navy's extramural projects demonstrated a 35% decrease (2,291) in animal use.

The increase in the Navy's intramural laboratory use of animals was in the area of infectious disease. Ninety-nine percent of the animals used by the Navy in infectious disease research were rodents. The majority of these animals were mice and were used in surveillance and research on new infectious diseases in the overseas research laboratories supporting the

Global Emerging Infectious Systems (GEIS) program. In the early 1990s, growing awareness and concern about the management of emerging infectious disease problems around the world led to meetings of public health experts sponsored by the National Academy of Sciences, by the World Health Organization/Pan American Health Organization, and by the White House. DoD representatives participated in these discussions. One result was a Presidential Decision Directive NSTC-7 in June 1996 that formally directed all federal agencies to cooperate in surveillance and research on new infectious disease problems. Because of its wide-ranging assets for disease control, the mission of the DoD was expanded to support global surveillance, training, research, and response to emerging infectious diseases. President Clinton directed a centrally coordinated program that improved DoD epidemiological capabilities and involved both U.S. military treatment facilities and military medical research units in the United States and abroad. The Navy's overseas laboratories represent 75% of the DoD overseas laboratory research program.

The Air Force used 4% of the total number of animals reported used by the DoD, 4% of the intramural animals, and 3% of the extramural animals. The Air Force had a 32% decrease (3,145) in intramural animal use and a 41% (2,541) decrease in extramural animal use resulting in a 36% (5,686) overall decrease in the number of animals used in research in FY98 compared with FY97. The Air

Force used the majority (6,681) of its animals in non-medical research projects.

The Office of the Secretary of Defense (OSD) components are the Uniformed Services University of the Health Sciences, Defense Advanced Research Projects Agency, Armed Forces Radiobiology Research Institute, and Armed Forces Institute of Pathology. OSD components used 10% of the DoD total animals used, 11% of the total intramural animals, and 9% of total extramural animals. There was a 39% (18,295) decrease in the use of animals for the OSD components in FY98 compared with FY97. This decrease was seen in both the intramural (6,740) and extramural (11,555) programs. The OSD components used the majority (92%) of their animals in clinical investigations (8,418) and medical research (18,538).

V.2.3 Animal Use by Species

The DoD has developed three major classifications for reporting animal use: non-mammals, rodents, and other mammals. Compared with FY97, in FY98 there was a 3% (700) decrease in the reported use of non-mammals and a 9% (26,230) decrease in the use of rodents (Figure V-6). However, there was a 28% (2,433) increase in the

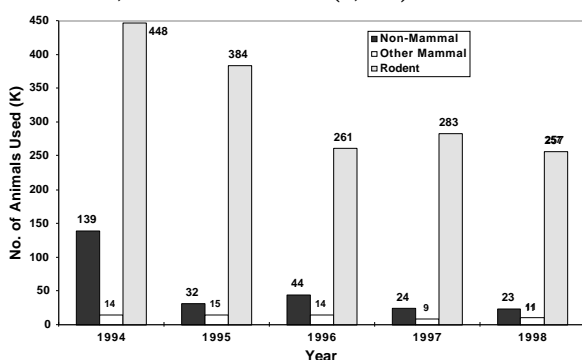


Figure V-6 Decrease in Species

number of other mammals used, with the bulk of this increase stemming from increases in the use of goats (1,432) and rabbits (1,022). During this same time period, there was a 6% (14,602) decrease in the use of mice, a 38% (13,931) decrease in rats, and a 35% decrease in amphibians (877). Still, the vast majority (96%) (280,426) of animals used by the DoD in FY98 were rodents, birds, amphibians, reptiles, and fish.

Since FY94, there have been significant decreases in the reported use of many species of

animals by the DoD. There has been an 83% (115,241) decrease in non-mammals, a 43% (191,040) decrease in rodents, and a 20% (2,696) decrease in other mammals. Several animals used in FY94 were not used at all in FY98 such as the fox, prairie dog, armadillo, civet, deer, and mink. In addition, there have been significant decreases in the use of large animals such as marine mammals and horses. For example, between FY94 and FY98 there was 59% (65) decrease in marine mammal and a 97% (102) decrease in horse use. At the same time, there were increases in the use of guinea pigs (4,098), swine (577), rabbit (135), chinchillas (73), gerbils (63), jirds (45), and ferrets (30). Most of the increase in guinea pig, goat, and rabbit use occurred between FY97 and FY98. Overall, there has been a shift from the use of large animals to smaller animals and a shift to those that are lower on the phylogenetic scale.

In FY98, there was a slight increase in the combined use of nonhuman primates, dogs, and cats. When comparing FY97 with FY98, there was an increase in the use of nonhuman primates (157) and a decrease in the use of dogs (33) and cats (21) (Figure V-7). Nonhuman primates were primarily used in medical research (81%), and within medical research, the majority (56%) of nonhuman primates were used in the area of infectious diseases.

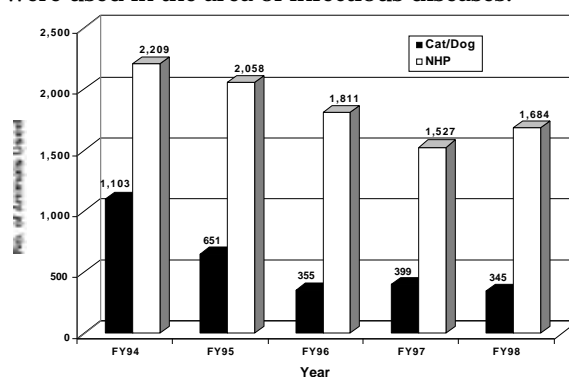
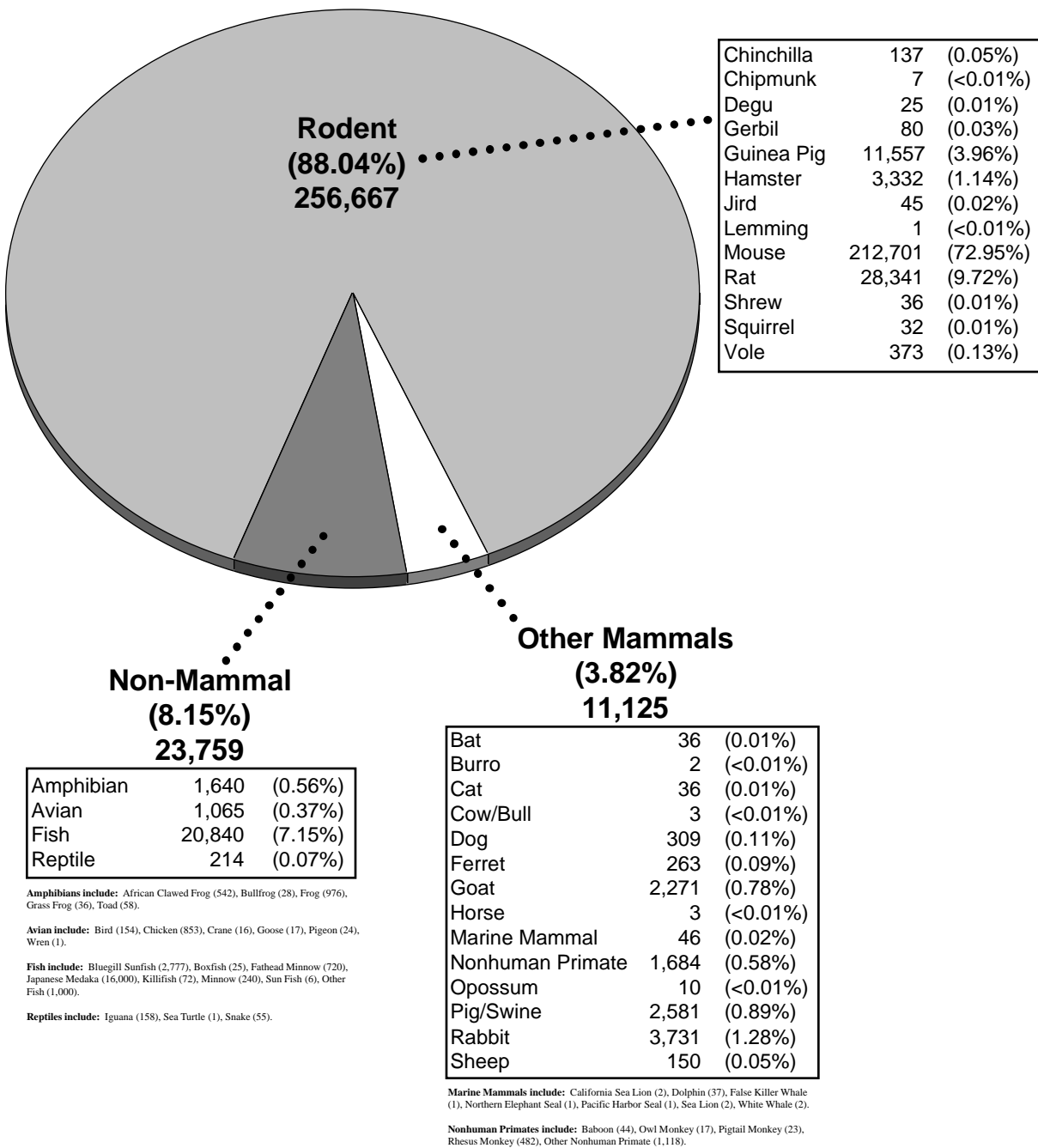


Figure V-7 Use of Nonhuman Primates, Dogs, and Cats by Year

Since FY94, there has been a 24% (525) decrease in the use of nonhuman primates and a 69% (758) decrease in the use of dogs and cats for research in the DoD. This illustrates the Department's continuing commitment to reducing the use of specific species in research.

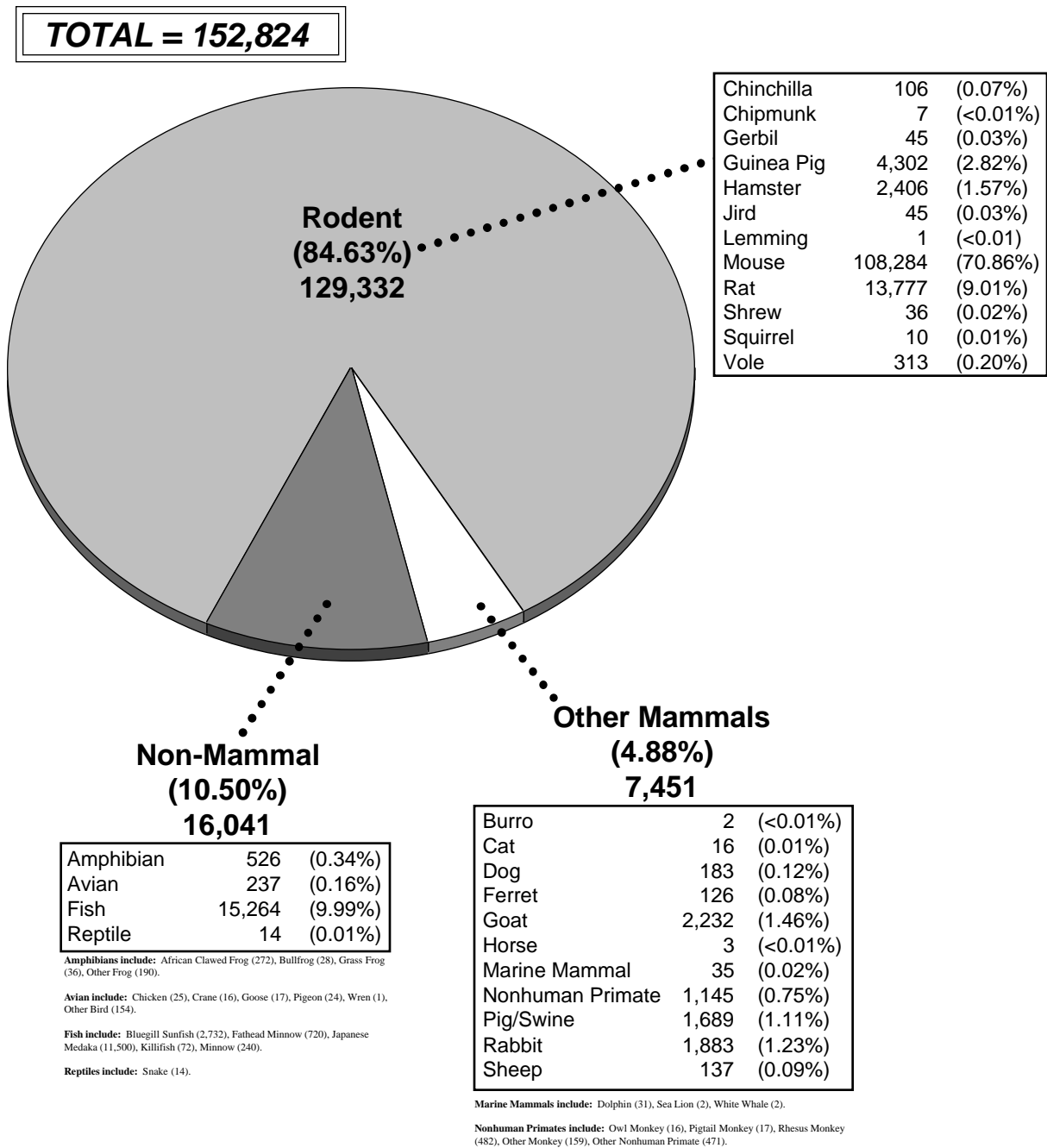
DoD animal use by species is presented in Figure V-8. Figures V-9 and V-10 represent the intramural and extramural animal use by species for FY98.

TOTAL = 291,551



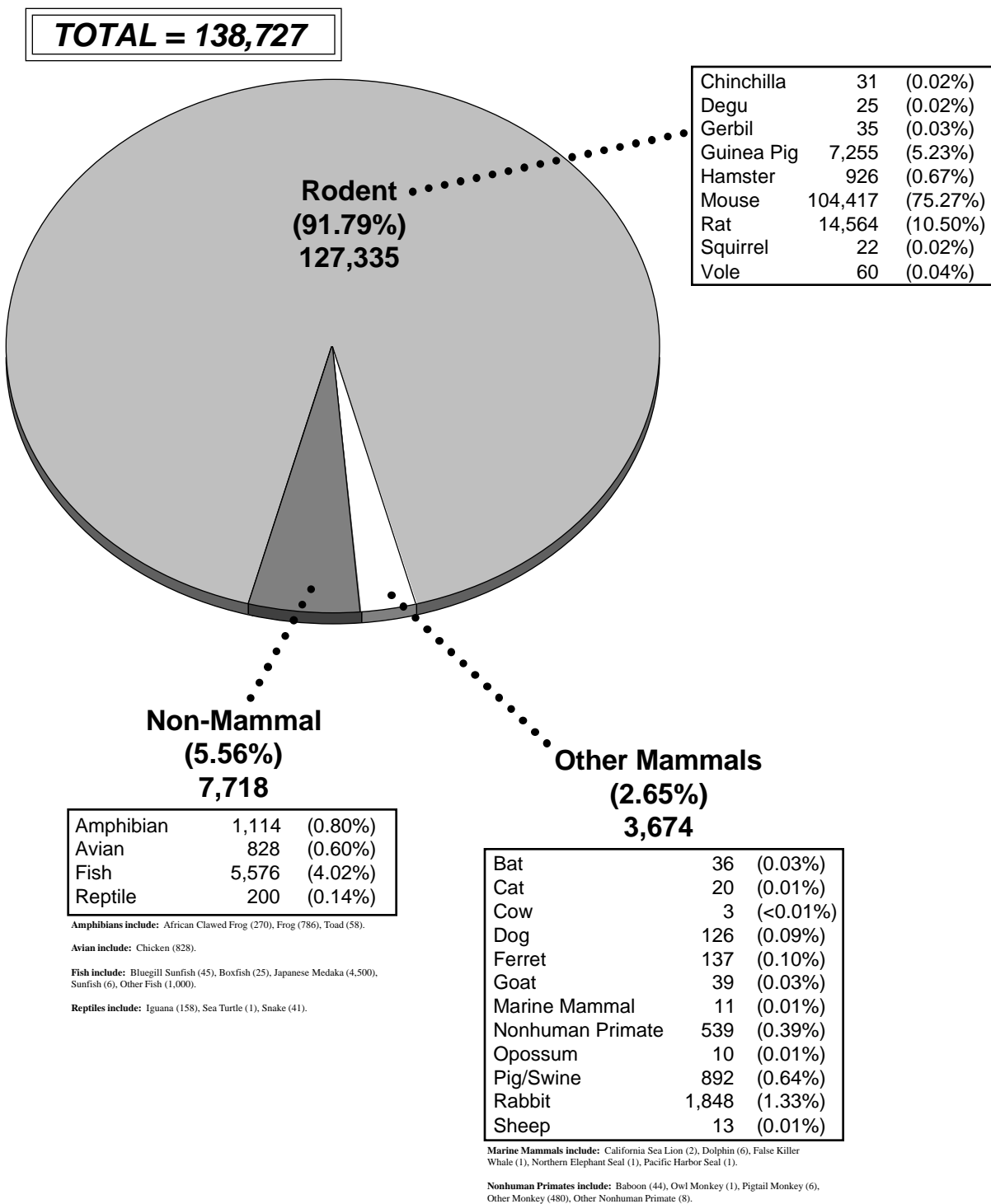
Percentages may not add up to 100% due to rounding of calculations

Figure V-8 DoD Intramural and Extramural Animal Use by Species for FY98



Percentages may not add up to 100% due to rounding of calculations

Figure V-9 DoD Intramural Animal Use by Species for FY98



Percentages may not add up to 100% due to rounding of calculations

Figure V-10 DoD Extramural Animal Use by Species for FY98

V.2.4 Animal Use by Category

Total reported animal use in the DoD by category is presented in [Figure V-11](#), with the intramural and extramural breakouts in [Figures V-12](#) and [V-13](#), respectively.

The DoD has a critical and challenging mission: to discover, design, and develop military medical countermeasures against threats to the health and survivability of military personnel. In order to meet this mission, 82% of the animals used by the DoD in FY98 were in medical research. Thirty percent (72,430) of the animals used in medical research were in the area of infectious diseases (M2) and of

those, 97% (69,333) were rodents ([Appendix O](#)). The primary thrust of this research is the development of preventive measures against infectious disease through discovery, design, and development of prophylactic, therapeutic, and treatment drugs for relevant diseases. The chemical defense research program (M3) used 16% (38,119) and the biological defense research program (M4) used 19% (46,403) of the medical research animals. Medical biological defense develops, demonstrates, and fields new vaccines, drugs, and diagnostic kits for the prevention, treatment, and diagnosis of biological warfare agents. This research program protects the armed forces from the consequences of exposure to biological warfare agents and enhances their

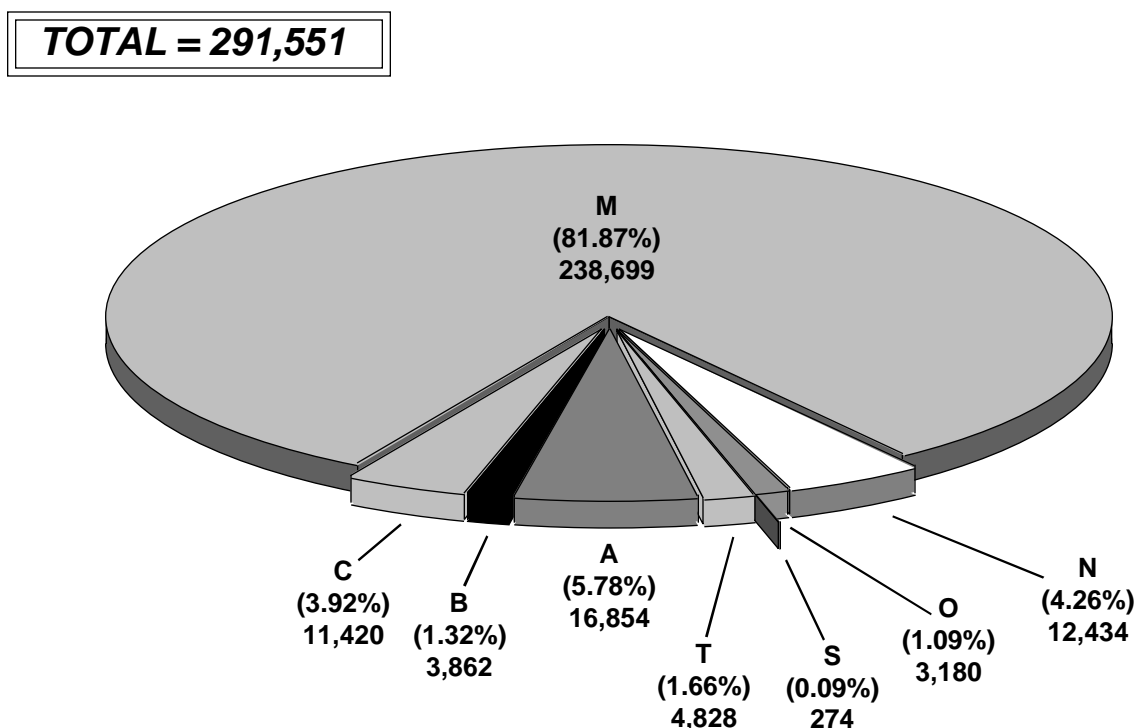


Figure V-11 DoD Intramural and Extramural Animal Use by Category for FY98

A: Adjuncts/Alternatives to Animal Studies, B: Animal Breeding Stock, C: Clinical Investigations, M: Medical RDT&E, N: Non-Medical RDT&E, O: Other Animal Use, S: Classified Secret or above, T: Training & Instructional.

Percentages may not add up to 100% due to rounding of calculations

TOTAL = 152,824

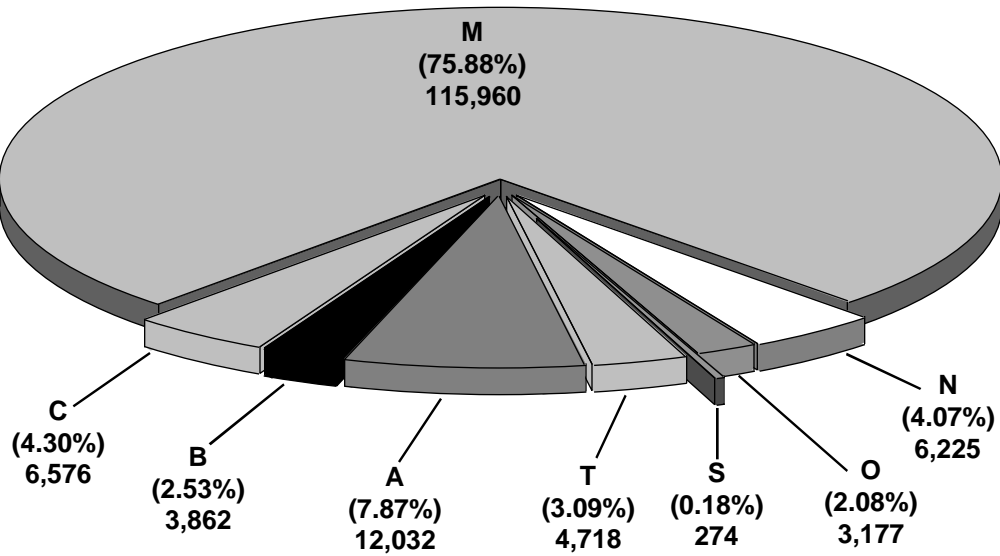


Figure V-12 DoD Intramural Animal Use by Category for FY98

TOTAL = 138,727

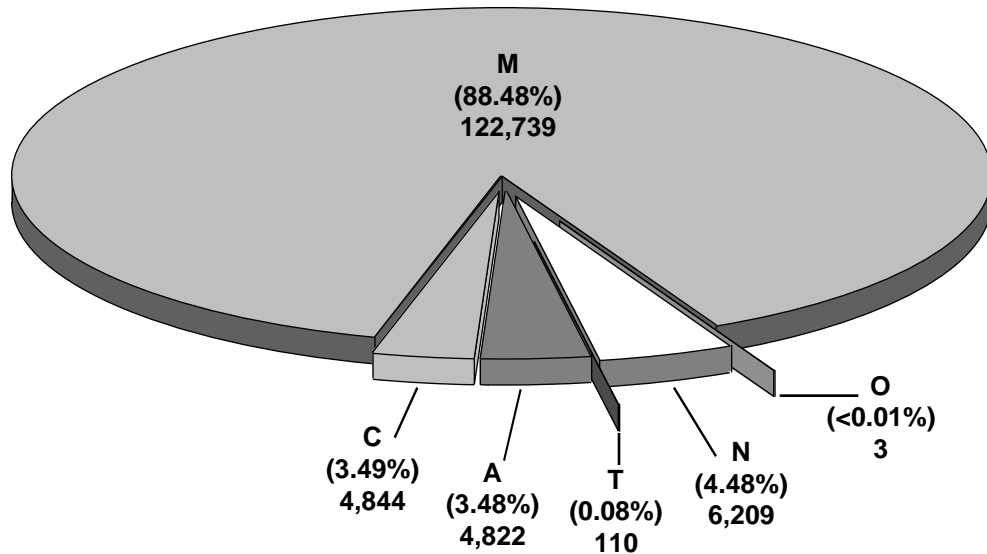


Figure V-13 DoD Extramural Animal Use by Category for FY98

A: Adjuncts/Alternatives to Animal Studies, B: Animal Breeding Stock, C: Clinical Investigations, M: Medical RDT&E, N: Non-Medical RDT&E, O: Other Animal Use, S: Classified Secret or above, T: Training & Instructional.

Percentages may not add up to 100% due to rounding of calculations

survivability. M8 (Other Medical Research) accounted for 25% of the total medical research category (Figure V-14). The Congressionally

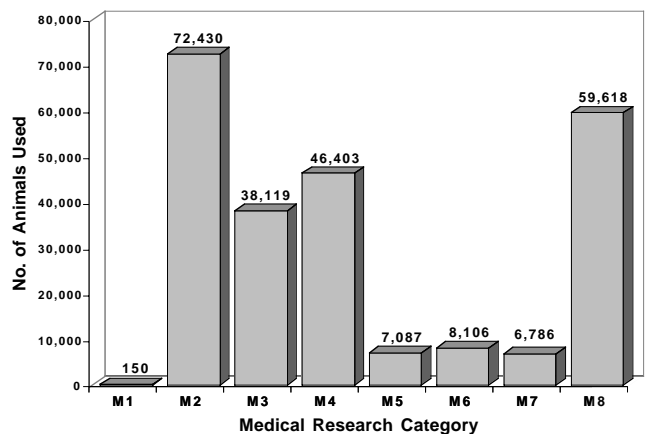


Figure V-14 Animal Use by Medical Research Category

Directed Research Programs in the areas of breast and prostate cancer, Defense women’s health, Gulf War illnesses, neurofibromatosis, and osteoporosis used 55,450 animals. These programs accounted for 93% of M8 animals (Table V-3), 23% of the animals used in medical research, and 19% of the total DoD animals used. These types of research programs can cause fluctuations in the total number of animals used from year to year depending on congressional funding levels and direction. Other areas of research within M8 are shown in Table V-3.

Research Category (M8)	No. of Animals Used	Percentage of M8
Biosample Protocol	5	0.01%
Breast Cancer	48,304	81.02%
Defense Women’s Health	141	0.24%
Disaster Relief & Emergency Medical Services	15	0.03%
Environmental Safety	30	0.05%
Gulf War Illnesses	1,167	1.96%
Medical Free Electron Laser	666	1.12%
Neurofibromatosis	2,539	4.26%
Neurotoxin	340	0.57%
Osteoporosis	404	0.68%
Prostate Cancer	2,895	4.86%
Toxicology	1,389	2.33%
Undersea Medicine	50	0.08%
Zoonosis	1,673	2.81%
Total M8 Research	59,618	100.00%

Table V-3 M8 (Other) Medical Research Category

Clinical research accounted for 4% (11,420) of the animals used by the DoD in FY98. Studies in this category address clinical medicine and surgical problems for the treatment of both diseases and

combat casualties. Ninety-one percent of the animals used in clinical research were used in clinical medicine studies. While many of these conditions are unique to the military, several are not. Specific types of clinical studies are listed in Appendix N.

Two percent of the animals used by the DoD in FY98 were in the training, education, and instruction of personnel. Training and instruction are basically for animal technicians and medical personnel (Appendix N). There was a 30% increase (1,126) in animals used in this category in FY98 compared with FY97. Breeding stock, classified studies, and other studies accounted for 3% of the DoD’s total animal use in FY98.

Non-medical RDT&E animal use decreased by 26% (4,347) in FY98 compared with FY97 and accounted for only 4% of the total animal use in FY98. Research in the area of alternatives to the use of animals increased by 39% (4,768) and accounted for 6% of the total animal use for FY98. Research in this category illustrates the Department’s continuing initiatives to promote research to develop alternatives to reduce, replace, and refine the use of animals in DoD research. No animals were reported as used for offensive weapons testing during FY98.

V.2.5 Animal Use by USDA Pain Category

Total reported animal use in the DoD by USDA pain category is presented in Figure V-15, with the intramural and extramural breakouts in Figures V-16 and V-17, respectively.

The majority (83%) of research in the DoD was not painful to the animals involved. In most cases (59%), the animals were not exposed to or involved in any painful procedures. In 24% of the cases, animals were given anesthesia or pain-relieving drugs during procedures that could have involved some pain or distress to the animals. In 17% of the animals used, anesthetics or analgesics were not used because they would have interfered with the validity of the results of experiments (Pain E category). There was a 22% decrease in Pain E animal use (13,751) between FY97and FY98. The majority of this decrease (9,312) was in the areas of medical chemical and biological defense research. Almost all (97%) of

the animals used in painful experiments (where reducing the pain or distress would have interfered with the validity of the results) were rodents. Only 1% of the animals in USDA Pain Category E were other mammals and less than 2% were non-mammals. Ninety-seven percent of the animals reported in USDA Pain Category E were used in medical studies; of these, 90% of the animals were used in research on infectious disease and chemical and biological defense. Infectious disease and chemical and biological defense research falls into USDA Pain Category E because the animals have to be exposed to chemical or biological agents or antidotes or other infectious diseases or vaccines, which may result in some type of distress. There were no animals subjected to unalleviated pain during training studies.

The DoD clearly has a most diverse, unique, and demanding R&D mission. The modern battlefield is a hostile and dangerous environment with extraordinary potential for exposure to lethal or debilitating conventional weapons, exotic

endemic diseases, biological and chemical agents, nuclear blast and radiation, directed energy sources, and complex and dangerous equipment. In addition, a host of adverse environmental conditions, such as cold, heat, high and low pressure, and G-forces are threats to service men and women. The DoD must provide acceptable protection against these threats and many others. The animals reported in USDA Category E were used in research designed to find ways to protect service men and women from the threats they encounter daily. Note that in most of these studies the distress level is minor, such as in heat stress or gastrointestinal distress after being exposed to G-forces. This critical research is often reliant upon animal models for vaccine and efficacious countermeasure development. Research of this kind is not commonly done elsewhere in the government, academic, or private sectors and therefore is the sole purview of the DoD. Also, a large portion of these studies are driven by federal requirements, particularly those of the Food and Drug Administration.

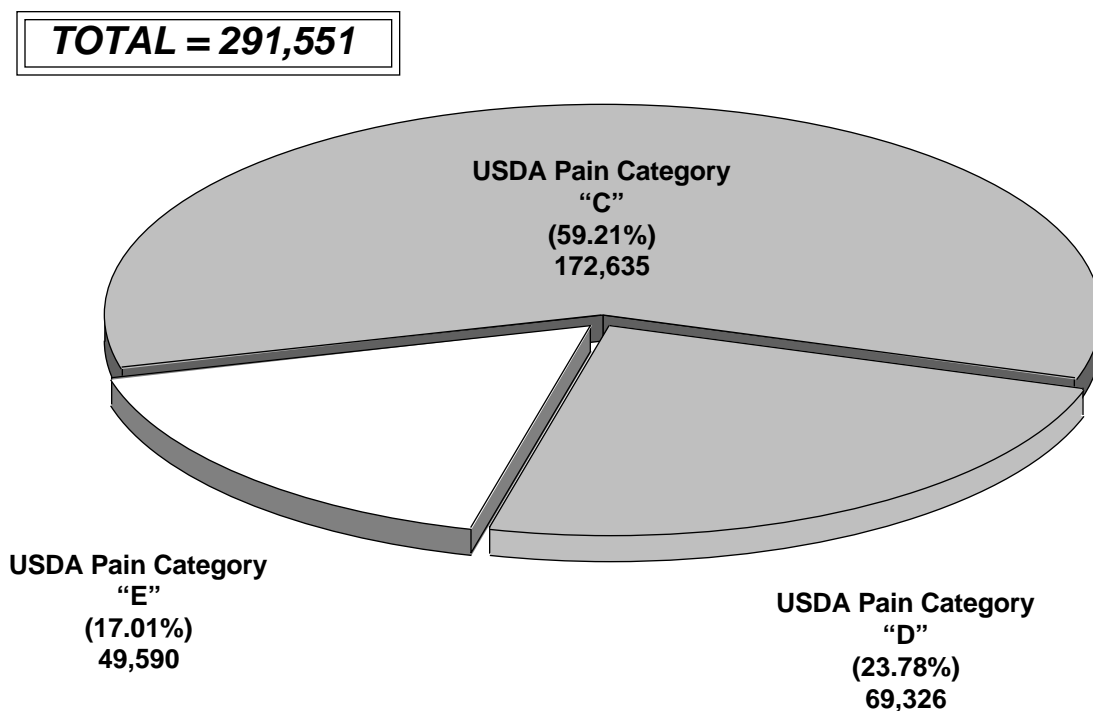


Figure V-15 DoD Intramural and Extramural Animal Use by USDA Pain Category for FY98

Percentages may not add up to 100% due to rounding of calculations

TOTAL = 152,824

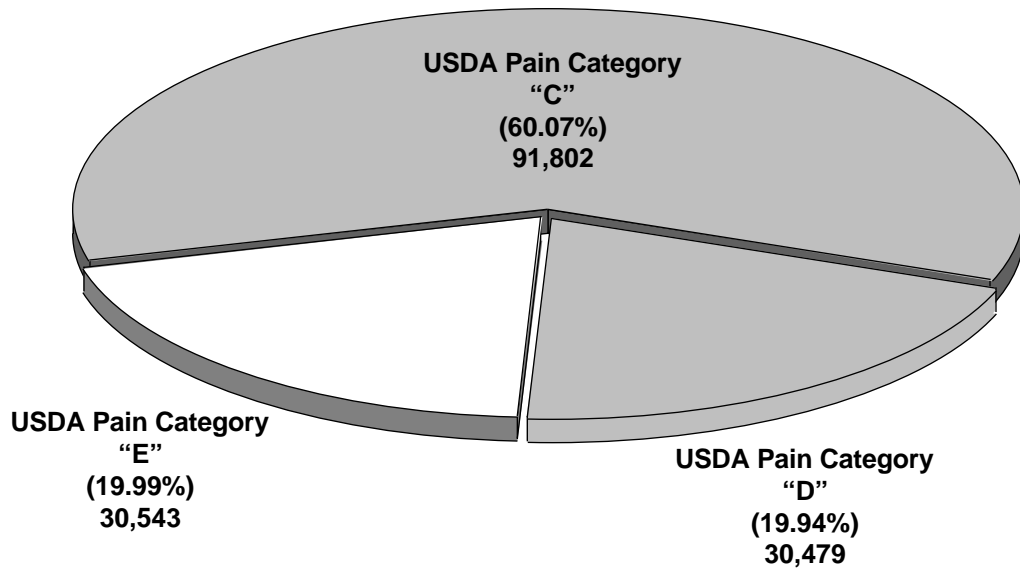


Figure V-16 DoD Intramural Animal Use by USDA Pain Category for FY98

Percentages may not add up to 100% due to rounding of calculations

TOTAL = 138,727

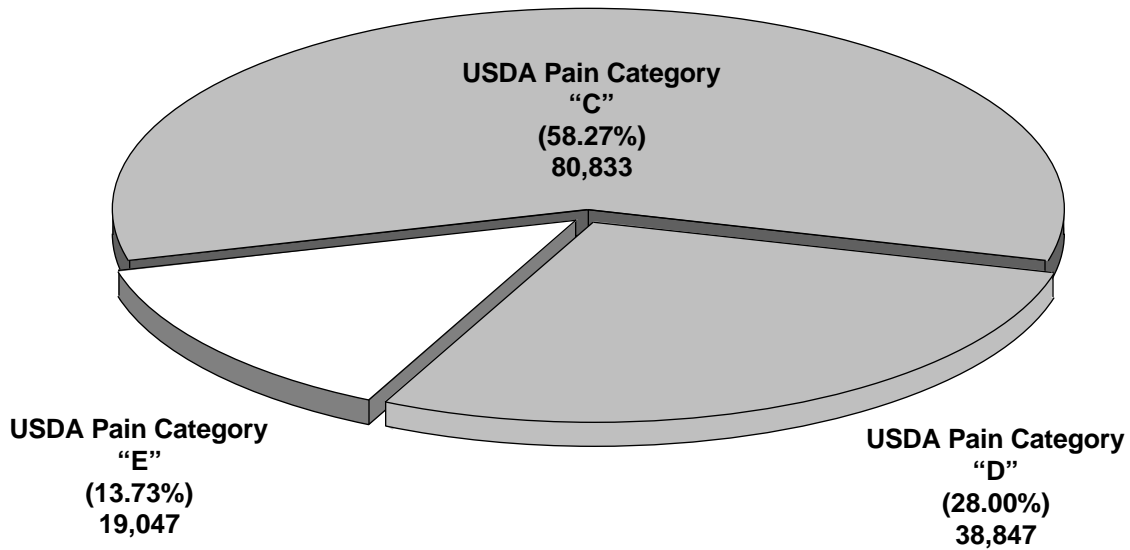


Figure V-17 DoD Extramural Animal Use by USDA Pain Category for FY98

Percentages may not add up to 100% due to rounding of calculations

SECTION VI

DoD INITIATIVES TO PROMOTE ALTERNATIVE METHODS THAT REPLACE, REDUCE, AND REFINES THE USE OF ANIMALS

Alternatives, as articulated in The Principles of Humane Experimental Technique (Russell and Burch, 1959), are defined as methods that Replace, Reduce, and Refine the use of animals. In addition to these Three Rs, the Department of Defense (DoD) advocates a fourth R, "Responsibility," for implementing these alternative methods.

Replacement

The replacement alternative addresses supplanting animal use with nonliving systems, analytical assays, cell-culture systems, and with animals that are lower on the phylogenetic scale. Additionally, human subjects are used when experimental drugs and other procedures progress to human trials. Such trials are conducted in accordance with Title 32, U.S. Code of Federal Regulations, Section 219, "Protection of Human Subjects in DoD-Sponsored Research."

Reduction

Decreasing the number of animals used through the use of statistical or innovative design strategies, while preserving the scientific integrity of the biological model, is a major emphasis of the reduction alternative to animal use.

Refinement

The refinement alternative for animal use addresses the need to ensure that the maximum humane use of each animal is obtained through proper protocol design and efficient utilization of animals, or through the modification of the experimental design to reduce the ethical cost associated with the study.

Responsibility

The DoD has taken responsibility for implementing animal use alternatives. This commitment illustrates the DoD's initiative toward utilization and development of alternatives to animal use.

Department policy with regard to animal alternatives is promulgated in DoD Directive 3216.1 which directs that "it is DoD policy that... alternatives to animal species should be used if they produce scientifically satisfactory results...." This policy is implemented in the Joint Service Regulation on the Use of Animals in DoD Programs, which delegates responsibility to the local commander for utilization of alternatives to animals.

To illustrate the Department's initiatives to promote these Four Rs, a description of such initiatives within DoD's research laboratories and medical treatment centers is provided. The following list is not all inclusive, as the number of specific examples of implementing alternative methods that can be documented for DoD's research projects is large. Rather, it illustrates the scope, diversity, and spirit of DoD's Four Rs initiatives. This section will demonstrate a broad-based movement toward the use of biotechnology and other innovative adjuncts to replace and reduce animal use as well as refinement in methods used in essential animal studies.

VI.1 DoD DEVELOPMENT OF ANIMAL USE ALTERNATIVES

A review of the FY98 DoD research reveals that 16 DoD organizations were actively involved in the development of alternatives to animal use. These developments occur through both research specifically designed to produce alternatives and by research to improve experimental techniques. Whenever possible, DoD investigators attempt to develop state-of-the-art, scientifically relevant and reliable experimental procedures that can be performed without the use of animals. In addition, in cases where the animal models cannot be completely replaced, investigators work diligently to develop refinement techniques to reduce any stress placed on the animal during both experimental procedures and daily living. The DoD is very active in the development of alternatives to

the use of animals in research. Below are examples of alternatives that the DoD reported to be finished with development in FY98. This is only a sample of the alternatives that completed development this year.

Replacement:

- A molecular assay was developed to replace suckling mouse assay in conjunction with DNA probes.
- The animal model for training in sinus surgery was replaced by virtual reality simulation.
- An artificial retina to be exposed to ultra short laser pulses to determine trends in damage threshold as a function of pulse width, wavelength, and number of pulses was developed.
- An artificial eye that mimics the focusing characteristics of the rhesus monkey eye was developed.
- A refined tissue culture system that replaces mice for ascites production in HIV research was developed.
- A reproductive toxicity test to replace laboratory mammals was developed using the gametes and embryos of *Xenopus laevis*.

Reduction:

- The propagation of a very unique polyclonal antibody was attempted in a small number of animals with long-term productivity in lieu of more animals with short production periods.
- In xenobiotic sensitization research, development of statistical and computer modeling systems reduced the number of rats used from 50 to 36.
- The number of mouse pups required for studying osteoblasts by freezing cells for long-term storage was reduced. When needed, frozen cells are thawed and expanded for use.
- In estrogen receptor research, methods were developed to harvest and store uterine tissues

at -80°C for several weeks, resulting in maximal use of the animals.

- In cellular analysis of the circadian clock, techniques were developed to examine individual neurons in culture, reducing the number of mice needed for experimentation.
- In studies on viral hemorrhagic and encephalitic diseases, the time period animals were left in the field was modified from 1 month to 3-6 months, thus reducing by two-thirds the total number of animals used.

Refinement:

- Determination of the shortest possible endpoint in research on reactions and interactions of neurons to nerve injury resulted in a reduction in animal days.
- Detailed postoperative discomfort monitoring and extended analgesic use were built into the protocol of knee reconstruction.
- Collection of ascitic fluid from the peritoneal cavity in mice was reduced from repeated collections to only one followed by immediate euthanasia in the evaluation of the potential threat of arboviral and other infectious diseases.
- In studies on experimental infection of dengue viruses, cynomolgus monkeys were caged in pairs to minimize distress.
- In special operations medical training, an operational and emergency training intubation was limited to five attempts per animal to minimize pain and distress.
- During the effect of immobilization on the healing of repaired achilles tendon, long-term analgesics were employed to avoid any possibility of pain and distress.
- Mice, carrying a transgene for spontaneous development of prostatic adenocarcinoma, were housed in sibling social groups with plastic "houses" to increase their well-being.

As an ongoing process, the DoD is continuously developing alternatives. Below are examples of

alternatives that were reported as currently in development by the DoD during FY98. This is only a sample of the alternatives being developed this year.

Replacement:

- Computer models for target detection to replace dolphins are being developed.
- Sonar and signal processing methods to replace marine mammals are in development.
- A mathematical method to understand the effect on human tissue of very high-peak-power electromagnetic pulses similar to those used in military electromagnetic weapons systems is being developed that will replace the use of animals in this research.
- Computer models of biologically relevant molecules have been developed so that one can test whether these molecules may be disturbed in some way by potentially hostile radiation environments.
- In research on maneuverability and performance versatility in swimming, a computer model is being developed to investigate fish swimming.
- A mouse model of apoptosis that will soon be replaced by tissue culture cells is in development.
- Eventual development of an "in vitro" correlate for anthrax will replace the use of animals for future studies.
- The use of mice in studying effects of drugs on tumor cells has been replaced by cell cultures.
- The validation of the susceptibility of rats to HEV infection may replace the use of monkeys.
- *Xenopus* is evaluated as an alternative sentinel species by studying the effects of reproductive system toxicants on reproductive organs and other endpoints.
- Medaka and bluegills are being evaluated as alternatives to rodents in studying the effect of

toxicants on immune system responses to foreign antigens.

- Medaka may replace laboratory rodents in studying the chronic toxicity of chemicals found in chlorinated drinking water.
- The development of rodent models will allow research on filoviruses to continue using animals lower on the phylogenetic tree than nonhuman primates.
- Validation of the guinea pig model will drastically reduce or eliminate the need for tests in nonhuman primates.
- Successful establishment of a murine model of infection will result in the replacement of hamsters as an experimental glanders model.
- Development of a murine model may replace nonhuman primates for screening of staphylococcal enterotoxin vaccine candidates.
- Implementation of tissue slices for the enhancement of drug development may replace the use of animals.

Reduction:

- A rabbit model for anthrax is under development that will result in a reduction in the number of nonhuman primates needed for future studies.
- The refinement of ribonucleic acid isolation procedures will reduce the number of animals used.
- In the study of rho-modifying cytotoxic necrotizing factor of *E. coli* in mice, the development of a tissue culture model will reduce the number of animals used.
- The use of organ culture to achieve gene suppression may provide an alternative to the generation and use of transgenic mice.
- Methods are currently being tested (polymerase chain reaction and immuno-staining) to determine if chiggers are infected with scrub typhus to reduce the number of rats that are needed for xenodiagnosis.

- The Multiplex Photonic/Electronic Sensor for biological warfare agents project is attempting to detect known and potential biological warfare agents without the use of animals to determine their pathogenicity, thereby reducing the number of animals needed to determine the probable pathogenicity of an agent.

Refinement:

- Digital dental radiographic equipment (Trophy system) offers the ability to develop multiple intraoperative radiographs quickly, as well as the ability to measure radiographic density through image analysis. Several types of data can thus be obtained simultaneously.
- Propagation of the Leishmania organism in the tail of the jird greatly reduces the apparent distress experienced by the subject.
- Determination of a surrogate marker to predict death instead of going to death as endpoint will reduce unnecessary stress.
- Development of methods of imaging retinal structure will reduce the need for histological examination of the fundus, which requires euthanasia.
- A method to launch ultra-low energy electromagnetic signals into an animal body to determine the animal's state of health is under development. This is a non-invasive physiological monitoring tool that permits biologists and physiologists to perform tests and measurements with diminished distress.
- Development of veterinary techniques training programs for authorized personnel utilizing various laboratory animal species will result in better animal handling.
- Various objects are being evaluated as potential sources of environmental enrichment for rabbits.
- Evaluation of a transdermal fentanyl system for swine may provide a much less stressful approach to analgesia and thus refine pain management.

- The possibility of noninvasively measuring internal body temperature through the use of diffusion magnetic resonance imaging to measure the temperature increase resulting from exposure to microwave fields is being investigated.
- Research on environmental enrichment for rhesus monkeys by engaging them in behavioral interaction that emulates the essential features of natural foraging is under way.

VI.2 DoD IMPLEMENTATION OF ANIMAL USE ALTERNATIVES

DoD research protocols strive to minimize the number of animals used to accomplish the program mission and goals. During the review of protocols by the Institutional Animal Care and Use Committee (IACUC), investigators are specifically asked to present information indicating that "Reduction, Replacement, and Refinement" have been addressed in the animal study. Implementation of these alternatives reduces, replaces, and refines the Department's use of animals in research. This is accomplished by the implementation of both general and specific alternatives. General alternatives are those that are frequently implemented in many different DoD programs. Specific alternatives are those that may be specific to both a research protocol and/or facility. In FY98, over 500 animal use projects reported that they were implementing alternative methods to the use of animals. There are too many general and specific alternatives implemented by the DoD to present all of them in this report. The following examples are a representative listing of general alternative methodologies commonly practiced in DoD facilities:

Replacement:

- During the review process, all potential methods of adequately answering the research objective are reviewed prior to the use of an animal model.

- The evaluation process also considers the selection of a particular animal type; species lower on the phylogenetic scale are considered and used if their selection permits attainment of the research objectives.
- Non-animal training aids are used to replace the use of live animals.
- Computer simulations are used to replace live animals when scientifically possible.

Reduction:

- All animal use protocols are subject to review by a biostatistician who addresses the animal used, study design, and statistical evaluation packages, and ensures that the minimum number of animals will be used to meet the specific scientific objectives.
- Pilot studies are used to refine techniques and define the animal model so that animal use can be kept to the minimum required for statistical significance.
- Sharing of animal tissues with other investigators reduces animal use.
- Iterations of the experiments will be combined when possible to reduce the number of control animals used.
- Collaboration between DoD investigators allows for a single animal to be used in multiple training and research procedures and the sharing of control group information, resulting in an overall reduction in the number of animals used.
- Several types of data are collected simultaneously.
- Training sessions are designed to use the highest practical student-to-animal ratio.
- When possible, animals serve as their own controls.
- Studies are deliberately phased so they continue progress only if warranted.

- Advanced experimental designs are developed that can reduce the number of animals used.

Refinement:

- Parameters indicating moribundity rather than death are used as experimental endpoints when possible.
- Animals are anesthetized during euthanasia to decrease stress.
- Moribund animals are humanely euthanized to prevent unnecessary pain or distress.
- Utilizing the environmental enrichment strategy, animals are housed in social settings (i.e., pairs or groups) in an enriched environment (e.g., nestboxes and toys).
- Basic animal handling skills and clinical techniques are taught to animal technicians, investigators, and research assistants to increase or ensure a proper skill level is attained prior to the start of a protocol.

Specific alternatives implemented by the DoD in FY98 were categorized as a subset of replacement, reduction, or refinement and are shown in [Table VI-1](#). These categories illustrate the broad-based spectrum of alternatives to be implemented by the DoD. A representative listing of the specific alternatives is presented in [Appendix P](#).

In addition to the implementation of alternatives, the DoD has established policies specific to the refinement of animal use. For example, Walter Reed Army Institute of Research (WRAIR) has established a policy that mandates consideration of environmental enrichment for research animals. This policy allows for flexibility and creativity for improving conditions of laboratory animals.

VI.3 DoD INITIATIVES TO PROMOTE ANIMAL ALTERNATIVES

The DoD has established a variety of initiatives and targeted programs that are currently in place to promote alternative methods that will replace,

Table VI-1 Alternatives Categories

<p>Replacement:</p> <ul style="list-style-type: none"> • Non-mammalian species or species lower in the phylogenetic scale • Biochemical/physical methods • Computer simulations • Other species replace companion animals • Replacement using in vitro cell cultures <p>Reduction:</p> <ul style="list-style-type: none"> • Utilization of alternative biological testing method • Substitution of computer simulations or other technologies • Substitution of another species • Changes in endpoint measurements <p>Refinement:</p> <ul style="list-style-type: none"> • Reduce pain • Reduce distress • Research models and animal alternatives • Environmental enrichment and improved animal handling
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reduce, and refine the use of animals. These programs are designed to target individual and institutional awareness by providing educational opportunities, professional training, and fiscal resources toward implementing the Four Rs approach to animal use.

VI.3.1 Science and Technology Objectives to Reduce Reliance on Animal Research

The DoD continues to seek alternatives to animal use through an Army Science and Technology Plan (STEP) initiated in FY93 and continuing through FY04 titled Reducing Reliance on Human and Animal Subjects of Research and Improving Experimental Conditions Using Animals. The objectives of the STEP are to conduct basic research to develop new technologies to incrementally reduce future reliance on research animals. The U.S. Army Medical Research and Materiel Command (USAMRMC) Medical Biological Defense Research Program budgeted approximately \$500,000 in FY98 for this objective, which is available to support alternatives to animal use research.

VI.3.2 DoD-Sponsored Conferences and Workshops on Alternatives to Animal Use

The DoD promotes responsibility for alternatives to animal use by sponsoring major meetings and conferences on the subject. Every 2 years the DoD sponsors an international meeting at Aberdeen Proving Ground on Alternatives to Animal Testing ([Table VI-2](#)).

Table VI-2 DoD-Sponsored Alternatives

Date	Title
1990	DoD Initiatives in Alternatives to Animal Testing
1992	Current Concepts and Approaches on Animal Test Alternatives
24-26 May 1994	Alternatives in the Assessment of Toxicity: Theory and Practice
12-14 June 1996	Biennial International Symposium on Alternatives in the Assessment of Toxicity Issues, Progress and Opportunities
December 1998	Alternative Toxicological Methods for the 21 st Century: Protecting the Human Health and Advancing Animal Welfare

The 1992 meeting had 35 scientific platform sessions and 22 scientific poster presentations. This international symposium was attended by nearly 300 military and civilian scientists from four countries. Proceedings of the 1992 symposium are available through the Defense Technical Information Center (DTIC). In addition, in 1994 a book edited by Dr. Harry Salem titled "Animal Test Alternatives" was published by Marcel Dekker, Inc., which included chapters prepared by most of the presenters at this symposium. The 1994 meeting had 26 scientific platform sessions,

including one by Dr. Martin Stephens of the Humane Society of the United States, and 45 scientific poster presentations. This meeting was attended by over 330 military and civilian scientists from seven countries. The proceedings and a monograph based on this successful symposium are available through DTIC. The book “Advances in Animal Alternatives for Safety and Efficacy Testing” was published by Taylor and Francis. The 1996 conference was coordinated with the Scientists Center for Animal Welfare, which held its meeting 10-11 June 1996 to present Animal Welfare and Toxicology/Safety Studies: Current Issues and Trends for the Next Century. In December 1998 the Alternative Toxicological Methods for the 21st Century: Protecting the Human Health and Advancing Animal Welfare conference was held in Bethesda, Maryland. This conference was sponsored by the Soldier and Biological, Chemical Command, The United States Army Center for Health Promotion and Preventive Medicine, the United States Army Institute for Chemical Defense and the National Institutes of Environmental Health Sciences. The purpose of this conference was to present the latest research and trends in programs to replace, reduce, or refine the use of research animals.

VI.3.3 National Research Council, Institute of Laboratory Animal Research, Educational Programs

The DoD’s priority and continuing commitment to promoting individual and institutional responsibility for alternatives to animal use are reflected in continuing financial support of the Institute of Laboratory Animal Research (ILAR) educational program of the National Research Council. The principal thrust of the ILAR grant is development of institutional training materials, educational courses, and publications in support of the Department’s laboratory animal care and use programs. This ILAR information is used in various military research facilities as an important adjunct to existing investigator training and technical education programs on animal care and use. The ILAR information and programs have generated strong animal alternative provisions for both civilian and military-specific research opportunities. The Department has funded this work since 1987 through 5-year grants, and is currently providing funding under the third such

grant. In the face of diminishing research funds, the Department has resolved to maintain this important collaboration by providing in excess of \$125,000 annually for the ILAR program.

VI.3.4 DoD’s Participation in Other Federal Alternatives Programs

The DoD is also represented on the Interagency Regulatory Alternatives Group (IRAG), which planned and presented a “Workshop on Updating Eye Irritation Test Methods” in 1991 and held another workshop on dermal testing held at the American College of Toxicology in November 1995. The DoD representative on the IRAG (Dr. Harry Salem) received the Food and Drug Administration’s (FDA’s) Group Recognition Award for his outstanding contributions to the IRAG ([Appendix Q](#)).

The National Institutes of Health Revitalization Act of 1993 (Public Law No. 103-43, Section 1301) directed the National Institute of Environmental Health Sciences of the National Institutes of Health (NIEHS/NIH) to establish an Applied Toxicological Research and Testing Program, which represents the NIEHS’ component of the National Toxicology Program. The Act further directed the NIEHS to “(a) establish criteria for the validation and regulatory acceptance of alternative testing methods, and (b) recommend a process through which scientifically validated alternative methods can be accepted for regulatory use.” To fulfill this mandate, an ad hoc Interagency Coordinating Committee on the Validation of Alternative Methods (ICC-VAM) was established in 1994 by NIEHS to develop a report recommending criteria and processes for validation and regulatory acceptance of toxicological testing methods that would be useful to federal agencies and the scientific community. The Department of Defense participated in this effort, which resulted in a report on the validation and regulatory acceptance of toxicological test methods.

Presentations have also been made on alternatives to the Board of Scientific Councilors of the National Toxicology Program of the National Institute of Environmental Health Sciences (NTP-NIEHS), Board of Scientific Councilors of the Food and Drug Administration, and Cancer Etiology Group at the National Cancer Institute.

VI.3.5 Institutional Animal Care and Use Committee Emphasis

Title 9 (Animals and Animal Products), Subchapter A (Animal Welfare), Parts 1-4 of the Code of Federal Regulations has specific provisions for addressing the issue of alternatives during the research animal protocol review process. The DoD has been a leader in forming lawfully constituted and functioning IACUCs at its biomedical research facilities. Accordingly, DoD IACUCs consider alternatives to the proposed use of animals as an important review consideration. All DoD programs use a standardized IACUC protocol format ([Appendix C](#)) for animal use proposals, which requires that non-animal alternatives be considered. It states that “No study using animals should be considered prior to the elimination of all reasonable possibilities that the question might be adequately answered using other than animal means.” Investigators must provide information on the animal model being proposed and justification for the selected species. The instructions for the standard protocol format states that “investigators should use the least sentient species that will permit the attainment of research objectives.” In addition, the investigators are required to provide a short description of the features of the proposal that may qualify the study as one that replaces, reduces, or refines the use of animals. The DoD 1995 Policy Memorandum ([Appendix B](#)) requires that extramural contractor proposals utilizing animals in research, testing, or training include all the information contained in the DoD standard protocol format, thereby requiring them to also provide the alternatives information.

VI.3.6 Veterinary Staff Expertise and Assistance Visits

The major biomedical research commands of the Military Departments each have credentialed laboratory animal medicine (LAM) veterinarians serving in key staff positions. More than 35 board-certified specialists of the American College of Laboratory Animal Medicine (ACLAM) currently serve in the DoD. In addition to being advisors to commanders on issues related to animal welfare and alternatives to animal use, these veterinarians provide oversight and structure to the Command's animal care and use programs. These officers also make periodic staff assistance visits to subordinate

facilities that use animals and evaluate each laboratory animal care and use program. Consideration of the use of alternatives is reviewed on these staff assistance visits. Another important responsibility of the LAM veterinarian is to review extramural animal use protocols, ensuring that alternatives to animal use and personnel training issues have been addressed.

VI.3.7 Professional Veterinary Training in LAM

The individuals who are specialty trained in veterinary LAM provide expertise in DoD biomedical research institutions, which strongly correlates to effective animal use alternatives programs. This is especially true in the critical area of refinements. The DoD has long been a leader in training veterinarians in the field of LAM, the biomedical and veterinary specialty most closely associated with laboratory animal welfare and laboratory animal care and use programs. Many of the nationally prominent leaders of several laboratory animal associations were formally trained in, or closely associated with, DoD LAM training programs. Examples are the President and several past presidents of ACLAM, the President-elect and several past presidents of the American Association of Laboratory Animal Science (AALAS), and several past presidents and the current Secretary-Treasurer of the American Society of Laboratory Animal Practitioners. This traditional DoD strength in LAM expertise strongly enhances both animal care and use and animal alternatives programs. Greater than 25% of all ACLAM boarded specialists in the U.S. received some or all of their LAM training in DoD LAM training programs.

VI.3.8 AALAS Technician and Laboratory Animal Science Training

There are a number of DoD research facilities that sponsor formal training programs leading to certification of animal care and research personnel as AALAS laboratory animal technicians. This specialized training is offered to both government and non-government animal technicians. It is an important mechanism for ensuring highly qualified animal care and research technicians in Defense laboratories. Individual DoD institutions have sponsored formal seminars for research personnel where experts from the National Agricultural

Workshop curriculum include formal training and information on alternatives to animal use. In addition, WRAIR offers quarterly a workshop on ethical and administrative issues relating to animal use. The AALAS technicians' course curriculum and the WRAIR workshop curriculum include formal training and information on alternatives to animal use.

VI.4 SUMMARY

Each year new techniques and capabilities improve the handling, treatment, and use of animals in research and testing, and potentially reduce the need for animals in those same endeavors. In FY98, there was ample evidence of the DoD's aggressive pursuit to develop alternatives to replace, reduce, and refine the use of animals (for example, the alternatives currently being developed and those that have finished development highlighted in [Section VI.1](#)). In

addition to these developmental efforts, animal use data for FY98 indicate the widespread implementation of validated alternatives. Rats and mice continued to replace nonhuman primates and other mammals higher on the phylogenetic scale in vaccine and drug development efforts. These and other examples of the development and implementation of alternatives have translated into reductions in the overall use of animals higher on the phylogenetic scale ([see Section V](#)). Animal use alternatives including reduction, replacement, and refinement constitute key initiatives in the biomedical research, testing, education, and training programs of the DoD. The number of large animals used by the military departments over the past decade has been significantly reduced, and some large species are rarely used at all. Dogs, cats, nonhuman primates, and marine mammals collectively represent less than 1% of the total animals used in research by the DoD.

SECTION VII

GLOSSARY

Adjuvant: An agent mixed in a vaccine to enhance the immunological protection afforded.

Alternatives to Animal Use: For purposes of this assessment, “alternatives” are defined as encompassing any subjects, protocols, or technologies that replace the use of laboratory animals altogether; reduce the number of animals required; or refine existing procedures or techniques so as to minimize the level of stress endured by the animal. These technologies involve the continued, but modified, use of animals; use of living systems; use of chemical and physical systems; and use of computers.

Analgesic: An agent that relieves pain without causing loss of consciousness.

Anesthetic: An agent that causes loss of the sensation of pain. Anesthetics may be classified as topical, local, or general.

Animal: For purposes of this assessment excluding embryos, animal is defined as any nonhuman member of five classes of vertebrates: mammals, birds, reptiles, amphibians, and fish. Within this group, two kinds of animals can be distinguished, warm-blooded animals (mammals and birds) and cold-blooded animals (reptiles, amphibians, and fish). Under this definition, invertebrates are not included.

Animal Use: The use of animals for research purposes. Three aspects of animal use are addressed in this assessment: behavioral and biomedical research; testing products for toxicity; and education of students at all levels. This assessment does not cover animal use for food and fiber; animal use to obtain biological products; or animal use for sport, entertainment, or companionship.

Animal Welfare Act: This act, passed in 1966 and amended in 1970, 1976, and 1985, was originally an endeavor to stop traffic in stolen animals that were being shipped across state lines and sold to

research laboratories. Amendments to the act have expanded its scope to include housing, feeding, transportation, and other aspects of animal care; however, the act bars regulation of the conduct of research and testing by the USDA.

Antibody: Proactive proteins produced by lymphocytes (a type of white blood cell) that can specifically bind foreign substances.

Association for Assessment and Accreditation of Laboratory Animal Care International (AAALAC): A voluntary private organization that has provided accreditation for over 600 institutions. AAALAC accreditation is based on the provisions of the National Research Council (NRC) *Guide for the Care and Use of Laboratory Animals*, and is recognized by the Public Health Service.

Biological Model: A surrogate or substitute for a process or organ of interest to an investigator. Animals or alternatives can serve as biological models.

Biomedical Research: A branch of research devoted to the understanding of life processes and the application of this knowledge to serve humans and animals. A major user of animals, biomedical research affects human health and the health care industry. It is instrumental in the development of medical products such as drugs and medical devices, and in the development of services such as surgical and diagnostic techniques. Biomedical research covers a broad spectrum of disciplines, such as anatomy, biochemistry, biology, endocrinology, genetics, immunology, nutrition, oncology, and toxicology.

Carcinogen: An agent or process that significantly increases the incidence of abnormal, invasive, or uncontrolled cell growth in a population. Carcinogens fall into three classes: chemicals, viruses, and ionizing radiation. A variety of screening assays have been developed to detect chemical carcinogens, including the *Salmonella*-

mediated mutagenesis assay (Ames test), the sister chromatid exchange assay, and traditional laboratory animal toxicity tests.

Cell Culture: Growth in the laboratory of cells isolated from multicellular organisms. Each culture is usually of one type. Cell culture may provide a promising alternative to animal experimentation, for example, in the testing of mutagenicity, and may also become a useful adjunct in repeated-dose toxicity testing.

Computer Simulations: The use of specially devised computer programs to simulate cells, tissues, fluids, organs, and organ systems for research purposes: to develop mathematical models and algorithms for use in toxicity testing, and to simulate experiments traditionally done with animals for educational purposes.

Distress: Usually the production of pain, anxiety, or fear. However, distress can also occur in the absence of pain. For example, an animal struggling in a restraint device may be free from pain, but may be in distress. Distress can be eased with tranquilizers.

Education: The aspect of education dealt with in this assessment is the use of animals and alternatives in the teaching of life sciences to health professionals and preprofessionals, and research scientists.

G-force: A unit measuring the inertial stress on a body undergoing rapid acceleration, expressed in multiples of the acceleration of gravity.

Guidelines for Animal Care and Use: Various organizations outside the federal government have adopted their own guidelines -- e.g., the American Psychological Association's *Guidelines for Ethical Conduct in the Care and Use of Animals*, which is comprehensive and has been endorsed by Federation of American Societies for Experimental Biology; the American Physiological Society's *Guiding Principles in the Care and Use of Animals*; and the American Veterinary Medical Association's *Animal Welfare Guiding Principles*. For federal guidelines, see Interagency Research Animal Committee, NRC *Guide for the Care and Use of Laboratory Animals*, and Public Health Service (PHS) Policy.

Institute of Laboratory Animal Research (ILAR): A component of the National Research Council, ILAR performs periodic surveys on the use of laboratory animals.

Institutional Animal Care and Use Committee (IACUC): An institutional committee that reviews research proposals and oversees housing and routine care of animals. The committee's membership generally includes the institution's attending veterinarian, a representative of the institution's administration, users of research animals, and one or more nonscientist and lay member.

Invertebrate: Any nonplant organism without a spinal column, e.g., worms, insects, and crustaceans. Invertebrates account for 90 percent of the earth's nonplant species. For the purposes of this assessment, invertebrates are not considered to be animals.

In vitro: Literally, in glass; pertaining to a biological process or reaction taking place in an artificial environment, usually a laboratory. Human and animal cells, tissues, and organs can be cultured in vitro. In vitro testing may hold some promising alternatives to animal testing, e.g., in testing for eye irritation and mutagenicity.

In vivo: Literally, in the living; pertaining to a biological process or reaction taking place in a living cell or organism.

National Research Council's *Guide for the Care and Use of Laboratory Animals*: Revised in 1996, the *Guide* details standards for animal care, maintenance, and housing. It is used by many animal research facilities, both within and outside the federal government. AAALAC and PHS also use it when assessing research facilities for accreditation.

Organ Culture: The attempt to isolate and maintain animal or human organs in in vitro culture. Long-term culture of whole organs is not generally feasible, but they can be sustained in cultures for short periods (hours or days).

Pain: Discomfort resulting from injury or disease. Pain can also be psychosomatic, the product of emotional stress. Pain can be induced by

mechanical, thermal, electrical, or chemical stimuli, and it can be relieved by analgesics or anesthetics.

Polymerase Chain Reaction: A molecular biological system in which pieces of genetic material can be synthesized in large amounts in vitro. This material can be used in diagnostic testing, genetic studies, or for a large number of molecular biological purposes.

Protocol: The written plan of a scientific experiment or treatment.

Public Health Service Policy on Humane Care and Use of Laboratory Animals: Revised in 1985, the *Policy* applies to PHS-supported activities involving animals [including those of the National Institutes of Health (NIH)]. It relied on the NIH *Guide for the Care and Use of Laboratory Animals* (1985), and uses institutional committees for the assessment of programs and maintenance of records.

Reduction: Considered an alternative to animal use when fewer animals are used in research and education through changed practices, sharing of animals, or better design of experimental protocols.

Refinement: An alternative to animal use by better use and modification of existing procedures so that animals are subjected to less pain and distress. Examples of such refinements are the administration of anesthetics and tranquilizers, humane destruction, and the use of noninvasive imaging techniques.

Replacement: An alternative to animal use, replacing methods using animals with those that do not. Examples include the use of a placenta

instead of a whole animal for microsurgical training, the use of cell cultures instead of mice and rats, the use of non-living systems, and the use of computer programs.

Research Facility: Under the Animal Welfare Act, any individual, institution, organization, or postsecondary school that uses or intends to use live animals in research, tests, or experiments. Facilities that receive no federal support for experimental work and that either purchase animals only within their own state or that maintain their own breeding colonies are not considered research facilities under the act.

Testing: Standardized procedures that have been demonstrated to predict certain health effects in humans and animals. Testing involves the frequent repetition of well-defined procedures with measurement of standardized biological endpoints. A given test may be used to evaluate many different substances and use many animals. Testing is used to establish the efficacy, safety, and toxicity of substances and procedures.

Tissue Culture: The maintenance in vitro of isolated pieces of a living organism. The various cell types are still arranged as they were in the original organism and their differential functions are intact.

Veterinary Medicine: The science and art of prevention, cure and/or alleviation of disease and injury in animals. Veterinary medicine includes the management of animal care and use programs.

SECTION VIII

REFERENCES

Army Science and Technology Master Plan, Fiscal Year 1997. Department of Army, March 1997

Department of Defense Directive 3216.1, "The Use of Laboratory Animals in DoD Programs," February 1, 1982; Revised, April 1995

Department of Defense Policy Memorandum, "Policy for Compliance with Federal Regulations and DoD Directives for the Care and Use of Laboratory Animals in DoD-Sponsored Programs," April 1995

Health Research Extension Act of 1985 (Public Law 99-158, November 20, 1995, "Animals in Research")

H.R. 96-1317, Department of Defense Appropriation Bill, 1981; Representative Addabbo, House Committee on Appropriations; 96th Congress, 2nd Session September 11, 1980

H.R. 97-332, Department of Defense Appropriation Bill, 1985; House Committee on Appropriation; 99th Congress, 1st Session October 24, 1985

Joint Regulation (Army Regulation 70-18; Secretary of the Navy Instruction 3900.38B; Air Force Regulation 169-2; Defense Advanced Research Projects Agency Instruction 18; Defense Nuclear Agency Instruction 3216.1B; Uniformed Services University of the Health Sciences Instruction 3203), "The Use of Animals in DoD Programs," June 1, 1984

National Academy of Sciences, National Research Council, Institute of Laboratory Animal Resources, *Guide for the Care and Use of Laboratory Animals*, 7th rev. edition, 1996

National Defense Authorization Act for Fiscal Year 1995, Report of the House Armed Services Committee, H.R. 4301, Report 103-499, May 10, 1994

President. 1996. Presidential Decision Directive NSTC-7. Subject: Emerging Infectious Diseases.

The National Institutes of Health Revitalization Act of 1993 (Public Law No. 103-43, Section 1301)

Public Health Service Policy on Humane Care and Use of Laboratory Animals

Report to the Committees on Armed Services of the Senate and House of Representatives on Department of Defense Animal Cost and Use Programs 1993

Report to the Senate Armed Services Committee and the House of Representatives National Security Committee on Department of Defense Animal Care and Use Programs 1994, 1995, 1996, and 1997

Review of Use of Animals in Department of Defense Contract Research Facilities, Inspector General, Department of Defense (August 1994)

Review of Use of Animals in the Department of Defense Medical Research Facilities, Inspector General, Department of Defense (February 1994)

Russell, W.M.S. and Burch, R.L., *The Principles of Humane Experimental Technique*, Charles C. Thomas Publishers, Springfield, IL, 1959

Title 7, United States Code, Sections 2131-2156, The Laboratory Animal Welfare Act of 1966, PL 89-544, as amended PL 94-279, 1976, and PL 99-198, 1985

Title 9, Code of Federal Regulations, Animals and Animal Products, Chapter 1: "Animal and Plant Health Inspection Service", Subchapter A: "Animal Welfare"; Source: 54 FR 36147, August 31, 1989

Title 32, U.S. Code of Federal Regulations Section 219, Protection of Human Subjects in DoD-Sponsored Research

Appendix A

DoD Directive on Animal Use



Department of Defense DIRECTIVE

April 17, 1995
NUMBER 3216.1

DDR&E

SUBJECT: Use of Laboratory Animals in DoD Programs

- References:
- (a) DoD Directive 3216.1, "Use of Animals in DoD Programs," February 1, 1982 (hereby canceled)
 - (b) Title 9, Code of Federal Regulations, "Animals and Animal Products," Chapter 1, Subchapter A, "Animal Welfare," Parts 1, 2, and 3
 - (c) Public Law 101-511, Department of Defense Appropriations Act for Fiscal Year 1991, Section 8019, Title 10 United States Code, Section 2241
 - (d) Sections 2131 through 2156 of Title 7, United States Code "The Laboratory Animal Welfare Act of 1966," as amended
 - (e) through (f), see enclosure 1.

A. REISSUANCE AND PURPOSE

1. Reissues reference (a) to update policy governing activities using animals within the Department of Defense.
2. Designates the Secretary of the Army as the DoD Executive Agent to develop and issue Service regulations to implement this Directive.

B. APPLICABILITY

This Directive applies to the Office of the Secretary of Defense, the Military Departments, the Uniformed Services University of the Health Sciences, and the Defense Agencies (hereafter referred to collectively as "DoD Components") that perform or sponsor activities using animals.

C. DEFINITIONS

Terms used in this Directive are defined in enclosure 2.

D. DoD POLICY

1. Federal statutes, regulations, and publications that provide national standards and guidance for the acquisition, transportation, housing, control, maintenance, handling, protection, treatment, care, use, and disposal of animals shall be applicable to all activities using animals. A summary of the applicable documents cited as references is in enclosure 3.

2. Animals shall be legally obtained from suppliers licensed by the U.S. Department of Agriculture (USDA) in accordance with

reference (b) unless specifically exempted from the licensing requirements stated in reference (b).

3. DoD organizations or facilities maintaining animals for use in research, testing or training shall apply for accreditation by the American Association for Accreditation of Laboratory Animal Care (AAALAC).

4. Alternative methods to animal species shall be considered, whenever possible, if such alternatives produce scientifically valid or equivalent results to attain the research testing and training objectives.

5. The purchase or use of dogs, cats, or nonhuman primates in research conducted for developing biological, chemical or nuclear weapons is prohibited.

6. The purchase or use of dogs, cats, or nonhuman primates for inflicting wounds from any type of weapon(s) to conduct training in surgical or other medical treatment procedures is prohibited. (reference (c)).

7. DoD organizations or facilities wishing to hold training programs using animals, such as advanced trauma life support (ATLS) training programs, shall have the training protocol reviewed and approved by a duly constituted Institutional Animal Care and Use Committee (IACUC) in accordance with references (d) and (e) and paragraph D.8. of this Directive to ensure the humane use of animals. DoD organizations or facilities conducting ATLS training that require housing of animals for short periods of time shall ensure adequate care and shall have the animal housing facilities inspected and approved by a veterinarian prior to receipt of the animals.

8. All proposals or protocols for animal experiments or demonstrations in RDT&E, clinical investigation, instructional, or training programs conducted or sponsored by a DoD organization or facility shall be reviewed and approved by a duly constituted IACUC composed of a minimum of five members. There shall be at least one non-scientific member on each IACUC. In addition, there also shall be a member who represents the general community interest and is non-affiliated with the facility sponsoring IACUC. The non-affiliated and the non-scientific membership can be filled by the same person. To ensure community representation at each meeting and inspection, an alternate to the non-affiliated member shall be designated for IACUCs having a single non-affiliated membership. Since the DoD IACUCs perform a Government function in an approval process and do not serve merely as an advisory body, the non-affiliated and the non-scientific member(s) to DoD IACUCs shall either be a Federal

employee, with demonstrated commitment to the community or a consultant consistent with the requirements established by reference (f).

9. A headquarters-level administrative review shall be conducted for proposals involving the use of non-human primates conducted or sponsored by subordinate activities of the DoD Component for conformance with all applicable Federal regulations and policies. A DoD component may delegate this responsibility to another DoD component for purposes of efficiency and consolidation of functional offices.

10. The DoD Components shall coordinate and cooperate in the transfer of Government-owned nonhuman primates between facilities to maximize conservation and proper utilization.

11. Proposals intending to use chimpanzees must be further reviewed and approved by the Interagency Animal Model Committee, which coordinates national priorities for research utilization of this species.

12. The DoD components that sponsor animal based research, testing, and training under a DoD grant or contract shall ensure that:

a. all extramural research proposals using live animals shall be administratively reviewed by a DoD veterinarian trained or experienced in laboratory animal science and medicine before grant or contract award.

b. the most recent USDA inspection reports are provided or obtained for the facility under consideration for a research contract or grant using animals, and that during the term of the award, the most recent USDA inspection reports be reviewed on an annual basis.

c. a DoD veterinarian trained or experienced in laboratory animal science and medicine shall conduct an initial site visit to evaluate animal care and use programs at contracted facilities conducting DoD-sponsored research using non-human primates, marine mammals, dogs, cats, or proposals deemed to warrant review. The initial site visit shall occur within 6 months of when the facility has taken delivery of the animals under DoD contract or grant award. Any facility receiving a DoD-funded grant or contract for animal based research shall notify the DoD component sponsor and shall have a site inspection within 30 days of notification of loss of AAALAC accreditation for cause, or notification that the facility is under USDA investigation. Site inspections for cause shall evaluate and

ensure the adequacy of animal care and use in DoD-sponsored programs, and provide recommendations to the sponsoring DoD component about continued funding support of the research.

13. In the case of differences between the standards of care and use of animals as cited in enclosure 3, the most stringent standard shall apply.

14. Activities covered by this Directive that are performed or sponsored in foreign countries shall be conducted in accordance with applicable U.S. statutory requirements, and regulations and standards of the host country. If differences exist between U.S. and host country regulations or standards, unless prohibited by the host country, the more stringent standard shall apply.

15. While not specifically addressed in this Directive, ceremonial, recreational, and working animals, such as military working dogs, shall be treated in a humane manner.

16. Personnel with complaints of violation of this directive shall report such violations to either of the following members of the organization or facility: IACUC chairperson, attending veterinarian, the facility Commander, or Inspector General. The IACUC shall review and, if warranted, investigate all reports of complaints of animal use or noncompliance with 7 U.S.C. 2131-2 of reference (d), applicable Directives, and regulations.

E. RESPONSIBILITIES

1. The Director, Defense Research and Engineering (under the Under Secretary of Defense for Acquisition and Technology) or designee shall:

a. Issue policy and procedural guidance concerning animal use consistent with all applicable Federal regulations and policies.

b. Designate a DoD representative to the Interagency Research Animal Committee who is a veterinarian of appropriate rank or grade and experience, and preferably also a diplomate of the American College of Laboratory Animal Medicine.

c. Establish the Joint Technical Working Group (JTWG) to act as the central advisory committee to the Armed Services Biomedical Research Evaluation and Management (ASBREM) Committee on all matters on the care and use of animals for research, testing, clinical investigation, or training within the Department of Defense. The co-chairpersons of the ASBREM Committee shall designate the chairperson of JTWG.

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2. The Heads of the DoD Components shall:

a. Establish appropriate mechanisms to monitor compliance with this Directive and applicable Federal statutes and regulations.

b. Establish offices or facilities that shall serve as reviewing or approving authorities of animal use proposals from subordinate activities and extramural facilities proposing research under contract or grant.

c. Provide members to JTWG as required.

d. Designate the appropriate office(s) within the DoD Component that shall perform the headquarters level administrative review of proposals requiring the use of non-human primates and shall serve as the office where exemptions under paragraph D.2. above may be approved.

e. Support, and as necessary, ensure the development of animal care and use training programs for researchers and members of the IACUC, and certification programs for all personnel involved in the care, use, and treatment of animals.

3. The Secretary of the Army shall:

a. As Executive Agent, develop and issue, in consultation with the other DoD Components, joint Service regulations to implement this Directive.

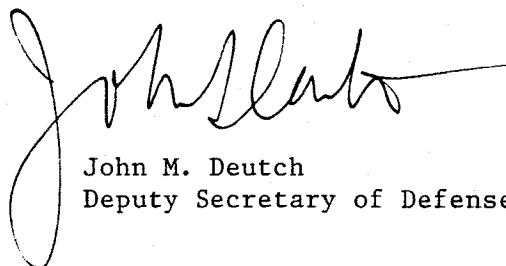
b. Designate the Commander, U.S. Army Veterinary Command/Director, DoD Veterinary Services Activity, a Field Operating Agency of the Army, Office of the Surgeon General who shall serve as a consultant to the Assistant Secretary of Defense for Health Affairs and the Director, Defense Research and Engineering for technical and professional matters related to this Directive.

F. EFFECTIVE DATE

This Directive is effective immediately.

Enclosures - 3

1. References
2. Definitions
3. Guidance Documents



John M. Deutch
Deputy Secretary of Defense

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- (e) National Institutes of Health (NIH) Publication No. 86-23, "Guide for the Care and Use of Laboratory Animals", United States Department of Health and Human Services, National Institutes of Health, Revised 1985.
- (f) Title 5, United States Code, Section 3109.

DEFINITION OF TERMS

1. Animal. - Any dog, cat, non-human primate, guinea pig, hamster, rabbit or any other live vertebrate animal, which is being used or is intended for use for research, training, testing, or experimentation purposes. For this Directive, it includes birds, rats of the genus *Rattus* and mice of the genus *Mus* bred for use in research, training, testing or experimentation purposes. The term excludes animals used for ceremonial or recreational purposes, military working animals, and animals intended for use as livestock and poultry as food or fiber; or, livestock or poultry used or intended for use for improving animal nutrition, breeding, management, or production efficiency, or for improving the quality of food or fiber.
2. Clinical Investigation. - All activities directed towards clinical research conducted principally within medical treatment facilities. The Clinical Investigations program is part of the Defense Health Program of the Assistant Secretary of Defense (Health Affairs) and is supported by Major Force Program 8 (MFP-8) funds.
3. Instructional Program. - All educational and training activities, except training of ceremonial and recreational animals and training associated with military working animals or survival skills training.
4. Research, Development, Test, and Evaluation. - All activities which form the RDT&E program of the Director, Defense Research and Engineering (DDR&E) and are supported by Major Force Program 6 (MFP-6) funds.
5. Alternatives. - Any system or method that covers one or more of the following: replacing or reducing the number of laboratory animals required for an investigation by computer simulation, cell culture techniques, etc; or, refining an existing procedure or technique to minimize the level of stress endured by the animal.
6. DoD Sponsored Programs. - All proposals or designs for animal experiments or demonstration in RDT&E, clinical investigation, or instructional programs conducted or funded by grant, award, loan, contract, or cooperative research and development agreement (CRADA).

**ADDITIONAL FEDERAL STATUTES, REGULATIONS,
AND GUIDELINES ON THE USE OF ANIMALS**

The following documents provide national standards and guidance for the protection, treatment and use of animals:

- a. **Animal Welfare Act** (Title 7, United States Code, Sections 2131-2158, as amended, and Title 9, Code of Federal Regulations, Parts 1-4, implementing rules and regulations). Administered by Regulatory Enforcement and Animal Care (REAC), Animal and Plant Health Inspection Service (APHIS) of the Department of Agriculture. Requires licensing of dealers, identification of animals, maintenance of records, submission of reports, establishment of an Institutional Animal Care and Use Committee (IACUC), and compliance with standards for the humane handling, care, treatment, and transportation of animals by dealers and research facilities.
- b. **Endangered Species Act of 1973** (Title 16, United States Code, Sections 1531-1543, as amended, and Title 50, Code of Federal Regulations, Parts 10-14 and 217-227, implementing rules and regulations). Provides a program under the U.S. Fish and Wildlife Service, Department of Interior, for conserving threatened and endangered species. Requires import/export permits, maintenance of records, and submission of reports on the care and handling of endangered, threatened, and conserved species.
- c. **Marine Mammal Protection Act** (Title 16, United States Code, Sections 1361-1384, as amended, and Title 50, Code of Federal Regulations, Parts 10-14 and 216-227, implementing rules and regulations). Provides a program under the Departments of Commerce (National Marine Fisheries Service) and Interior (U.S. Fish and Wildlife Service) for the protection of marine mammals and marine mammal products. Requires acquisition permits, maintenance of records, submission of reports, and inspections on the care and handling of marine mammals.
- d. **Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES)** (TIAS 8249, as amended, and Title 50, Code of Federal Regulations, Part 23, implementing rules and regulations). CITES is a treaty involving 106 signatory nations administered in the United States by the Fish and Wildlife Service of the Department of the Interior. CITES regulates the import and export of imperiled species covered by the treaty but imposes no restrictions or control on interstate shipments.
- e. **Lacey Act** (Title 18, United States Code, Section 42, as

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amended, and Title 50, Code of Federal Regulations, Part 16 and Subpart B, implementing rules and regulations). A program under the U.S. Fish and Wildlife Service, Department of the Interior. Prohibits the importation of certain wild animals or their eggs if the Secretary of the Interior determines that they are injurious to humans, the interest of agriculture, or other specified national interests.

f. **Guide for the Care and Use of Laboratory Animals.** Public Health Service, National Institutes of Health, NIH Publication No. 86-23, Revised. Provides guidelines for institutional policies, husbandry, requirements, veterinary care, and physical plant requirements for programs involving the care and use of laboratory animals.

g. **Guide for the Care and Use of Agricultural Animals in Agricultural Research and Teaching.** Published by the Consortium for Developing a Guide for the Care and Use of Agricultural Animals in Agricultural Research and Teaching, 309 West Clark Street, Champaign, IL 61820, March 1988. Provides guidelines for the care and use of the major agricultural animal species in the United States in research and teaching.

Appendix B

**Department of Defense (DoD) Policy for Compliance with Federal
Regulations and DoD Directives for the Care and Use of
Laboratory Animals in DoD-Sponsored Programs**



OFFICE OF THE SECRETARY OF DEFENSE

WASHINGTON, D.C. 20301

10 APR 1995

MEMORANDUM FOR ASSISTANT SECRETARY OF THE ARMY (M&RA)
ASSISTANT SECRETARY OF THE ARMY (RDA)
ASSISTANT SECRETARY OF THE NAVY (M&RA)
ASSISTANT SECRETARY OF THE NAVY (RDA)
ASSISTANT SECRETARY OF THE AIR FORCE
(MRAI&E)
ASSISTANT SECRETARY OF THE AIR FORCE (SAF/AQ)
PRESIDENT, UNIFORMED SERVICES UNIVERSITY OF THE
HEALTH SCIENCES
DIRECTOR, DEFENSE NUCLEAR AGENCY
DIRECTOR, ADVANCED RESEARCH PROJECTS AGENCY

SUBJECT: Department of Defense (DoD) Policy for Compliance with
Federal Regulations and DoD Directives for the Care and
Use of Laboratory Animals in DoD-Sponsored Programs

References:

(a) Title 7, United States Code, Sections 2131-2156,
The Laboratory Animal Welfare Act of 1966, PL 89-544,
as amended PL 94-279, 1976, and PL 99-198, 1985.

(b) Review of the Use of Animals in the Department of
Defense Medical Research Facilities, Inspector General
Department of Defense, February 1994.

(c) Review of the Use of Animals in Department of
Defense Contract Research Facilities, Inspector
General Department of Defense, August 1994.

Definition:

(a) Animal means any dog, cat, non-human primate, or
any other live vertebrate animal which is being used
or is intended for use for research, training, testing,
or experimentation purposes. For this Policy Guidance,
it includes birds, rats of the genus Rattus and mice of
the genus Mus bred for use in research, training,
testing or experimentation purposes. The term excludes
animals used for ceremonial or recreational purposes,
military working animals, and animals intended for use
as livestock and poultry as food or fiber; or,
livestock or poultry used or intended for use for
improving animal nutrition, breeding, management, or
production efficiency, or for improving the quality of
food or fiber.

(b) DoD-Sponsored programs means any study, proposal,
or design for animal experimentation or demonstration
in Research Development, Test, and Evaluation (RDT&E),
clinical investigation, or instructional program
conducted or funded by grant, award, loan, contract, or
cooperative research and development agreement (CRADA).

Reference (a) has been accepted by the Department of Defense (DoD) in the development of DoD Directives and policy guidance. References (b) and (c) contain recommendations which have been endorsed by the Department. The purpose of this policy memorandum is to implement the recommendations contained in references (b) and (c).

DoD components that utilize animals in DoD-supported programs shall be aware of the attached DoD Directive 3216.1, "Use of Laboratory Animals in DoD Programs," appended as attachment (1). It is currently pending signature and will supersede the current DoD Directive 3216.1 dated February 1, 1982. Additional policy guidance is as follows:

a) In DoD component facilities conducting animal-based programs, an alternate to the non-affiliated member of the Institutional Animal Care and Use Committee (IACUC) shall be designated for IACUCs having a single non-affiliated member. The non-affiliated member(s) or alternates must receive a minimum of eight hours training. At least four hours of the training shall address the regulatory responsibilities and proper techniques on animal protocol review processes. An additional minimum of four hours of training will address humane care and ethics issues dealing with animal use. All DoD Components conducting animal use programs as defined shall have training programs for non-affiliated IACUC members in place by 1 October 1995.

b) All DoD component facilities maintaining animals used in research, testing, or training shall apply for accreditation by the American Association for the Accreditation of Laboratory Animal Care (AAALAC). The Office of the Director, Environmental and Life Sciences, Pentagon Room 3D129, Washington, D.C. 20301-3030 is the central point of contact to maintain cognizance over the application or continuation of AAALAC accreditation. All DoD facilities shall furnish copies of AAALAC accreditation status to that office. Absence of accreditation shall be explained with a plan of action and milestones to obtain accreditation.


The following recommendations from the DoD Inspector General have been adopted as policy and shall be fully implemented by DoD Components which use animals in DoD-sponsored programs.

a) The DoD standard protocol format appended as attachment (2) shall be implemented by 1 October 1995. All intramural protocols involving animal use submitted after 1 October 1995 shall use the standard format. Extramural contractor proposal submissions need not use the standard format; however, the contractor shall provide all pertinent information contained in the standardized protocol format.

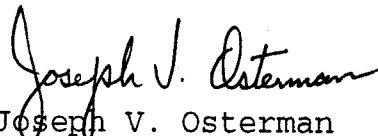
b) All DoD component facilities that utilize animals in research, testing and training shall implement the DoD standardized semi-annual program review checklist appended as attachment (3) immediately. Accompanying the checklist is a detailed outline of program review as contained in the NIH Guide for the Care and Use of Laboratory Animals. The Guide is the primary reference which is used by AAALAC in the accreditation process. The checklist shall be completed as a part of the semiannual IACUC program and facility review process. The semi-annual IACUC reports shall contain a copy of the checklist or indicate that the checklist was used as the basis of the program and facility review. A majority of members of the IACUC shall sign the report and include a statement indicating the presence or absence of minority opinions.

c) Commanders, and Directors of DoD component facilities shall support and, as necessary, develop animal care and use training programs for personnel associated with animal use programs, and encourage certification for all personnel involved in the care, use and treatment of laboratory animals.

As of 1 October 1995, DoD components shall report all animal-based protocols in the required format redacted for public release to the Defense Technical Information Center (DTIC). Selected fields of the DTIC report will be made accessible to the public through the INTERNET.



Edward D. Martin
Principal Deputy,
Assistant Secretary of
Defense (Health Affairs)



Joseph V. Osterman
Director, Environmental
and Life Sciences

Attachments:

- (1) Pending DoD Directive 3216.1
- (2) Standard Protocol Format
- (3) Standard Semi-annual Checklist

Appendix C

DoD Standard IACUC Protocol Format Instructions

ALL DOD ANIMAL USE PROTOCOLS MUST UTILIZE THIS DOD STANDARDIZED FORMAT. This protocol format only includes those requirements of the Animals Welfare Act, American Association for the Accreditation of Laboratory Animal Care, Federal Regulations, DoD Directives and DoD Policy relating to animal use. Any requirements that are specific to a given Service, Command, or locale (such as all budgeting information, local coordinating requirements, specific scientific review requirements etc.) should be added by each organization in front or behind this standardized format. Adding some information within the format is acceptable to meet local needs as long as the standard format is maintained. In other words, all of the labelled paragraphs and subparagraphs should remain in the same relative order with the added information being similar or complementary to the information requested. It is important to note that this standardized protocol format does not in any way prohibit local organizations from using any (or all) of their current animal use protocol. It does mandate that all of the information required in this DoD standardized format be answered as a part of the organization's animal use protocol in the order listed in this format.

THIS DOCUMENT IS INTENDED TO BE AN AID IN THE PREPARATION OF A DOD ANIMAL USE PROPOSAL. IT IS A COMPANION DOCUMENT TO AN IDENTICAL PROTOCOL FORMAT OR TEMPLATE THAT DOES NOT HAVE THE WRITTEN EXPLANATION FOR INDIVIDUAL PARAGRAPHS. THEY ARE DESIGNED TO BE USED ON A WORD PROCESSING PROGRAM, i.e., WordPerfect, WordStar, MicrosoftWord, WordPerfect for Macintosh, etc., SO THAT YOU ARE NOT LIMITED BY THE SPACE PROVIDED, AND SUGGESTED CHANGES OR MODIFICATIONS CAN BE QUICKLY AND EASILY MADE. USING A WORD PROCESSOR MAKES THIS FORMAT A "FILL-IN-THE-BLANKS" EXERCISE. THE EXPLANATIONS OR INSTRUCTIONS MAY BE BLOCKED OUT AND DELETED IF IT IS MORE CONVENIENT TO USE THIS FORM RATHER THAN THE OUTLINE AVAILABLE WITHOUT THE EXPLANATIONS. SPECIFIC RESPONSES REQUESTED IN THE FORMAT ARE A RESULT OF THE REQUIREMENTS OF THE ANIMAL WELFARE ACT (AWA), DOD REGULATIONS, OR ANIMAL WELFARE GUIDELINES. EACH PARAGRAPH SHOULD HAVE A RESPONSE. PORTIONS OF THE PROTOCOL FORMAT THAT ARE NOT APPLICABLE TO YOUR PARTICULAR PROTOCOL, i.e., NO SURGERY OR NO PROLONGED RESTRAINT, SHOULD BE MARKED N/A. IF SOPs OR OTHER DOCUMENTS ARE READILY AVAILABLE TO THE IACUC, THEY MAY BE REFERENCED TO ASSIST IN THE DESCRIPTION OF SPECIFIC PROCEDURES. IT IS CRITICAL THAT ONLY ANIMAL STUDIES OR PROCEDURES DOCUMENTED IN AN APPROVED PROTOCOL ARE PERFORMED IN THE ORGANIZATION. ADDITIONALLY, P.I.s OR OTHER ANIMAL USERS SHOULD KEEP ACCURATE EXPERIMENTAL RECORDS, AND BE ABLE TO PROVIDE AN AUDIT TRAIL OF THEIR ANIMAL EXPENDITURES AND USE THAT CORRELATES TO APPROVED PROTOCOLS.

PROTOCOL COVER SHEET: Requires a minimum of three signatures to include: the Primary Investigator, the individual responsible for scientific review and the Attending Veterinarian. In addition, the signature from the individual performing the statistical review on this cover sheet is recommended. If no signature block is present for a person who does the statistical review, then the following statement must be present on the protocol cover sheet. "A person knowledgeable in statistics has reviewed the experimental design." This Protocol Cover Sheet can also hold any additional information deemed necessary by the organization (Co- investigators, Department/Division Chief, Coordinating Departments, IACUC Chair, Biosafety Review etc.)

PROTOCOL TITLE:

PRINCIPAL INVESTIGATOR:

(Signature Required)

(Principal Investigator)

SCIENTIFIC REVIEW: Signature verifies that this proposed animal use protocol has received appropriate peer scientific review, and is consistent with good scientific research practice. (No response is required to the title paragraph of this section)

(Signature Required)

(Research Unit Chief/Directors signature)

ATTENDING/CONSULTING VETERINARIAN: (Example) The attending/consulting veterinarian has reviewed the protocol and was consulted in the planning of procedures that require veterinary input, i.e., an unalleviated pain procedure. In addition, the veterinarian/veterinary medicine department has assisted with coordination for veterinary support to the protocol. (No response is required to the title paragraph of this section)

(Signature Required)

(Attending/Consulting Veterinarian)

STATISTICAL REVIEW: A person knowledgeable in statistics has reviewed the experimental design. (No response is required to the title paragraph of this section) (Inclusion of Signature Block is Recommended, but Optional)

(Statistician)

OTHERS: You may wish to add specific additional offices or signature blocks for individuals responsible for coordination or compliance issues pertinent to your facility or operation. (i.e. Co-investigators, Coordinating Departments, IACUC Chair, Biosafety Review etc.)

PROTOCOL TITLE:

PRINCIPAL INVESTIGATOR:

CO-INVESTIGATOR(S):

I. NON-TECHNICAL SYNOPSIS: A brief, narrative description of the proposal or idea that is easily understood by non-scientists.

II. BACKGROUND:

A. Background: This should include a brief statement of the requirement or need for the information being sought. Lengthy explanations are not required. Typically, the "literature or the experience that led to the proposal will be briefly reviewed" (AR 70-18), and a description of the general approach should be provided. Unnecessary duplication of effort should be strictly avoided.

B. Literature Search: This search must be performed to prevent unnecessary duplication of previous experiments. A search of Federal Research in Progress (FEDRIP) and DTIC databases or their equivalent is required for DOD funded research. An additional search of the scientific literature (MEDLINE, GRATEFUL MED, MEDLARS, AWIC, etc.) is highly recommended.

1. Literature Source(s) Searched:

2. Date and Number of Search:

3. Key Words of Search:

4. Results of Search: Provide a narrative description of the results of the literature search(s).

III. OBJECTIVE\HYPOTHESIS: In non-technical terms, state the objective of this protocol, or the hypothesis to be accepted or rejected.

IV. MILITARY RELEVANCE: With regards to military needs and mission requirements, this paragraph should provide a brief and succinct military justification for the research. If applicable state the Science and Technology Objective (STO) that this work supports.

V. MATERIALS AND METHODS:

A. Experimental Design and General Procedures: Provide a "complete description of the proposed use of animals." This section should succinctly outline the formal scientific plan and direction for experimentation. If several experiments or sequential studies

are to be included in the protocol, description of the experimental design for each separate experiment should be contained in sub-parts to this section. The length and detail required in this section depends largely on the complexity of the study. However, **a clearly understandable description of the numbers of animals and their distribution into experimental groups is essential.** The number requested should be the minimum numbers necessary to complete the study, but must be sufficient to yield meaningful results. If too few animals are requested and statistical significance is not achieved, the animals will have been misused. Be certain to include animals necessary for controls or technique development, etc. If the design is complex, a summary table or flow chart showing the distribution of animals by experimental group should be included. **The total number of animals required for the study is listed in section V.B.4. It is critical that reviewers of this protocol are able to follow your reasoning and calculations for the number of animals required, and can verify that the experimental design clearly supports the number of animals requested.**

1. Experiment 1:

2. Experiment 2: (etc.)

B. Laboratory Animals Required and Justification:

1. **Non-animal Alternatives Considered:** Were alternatives to animal use considered? **No study using animals should be considered prior to the elimination of all reasonable possibilities that the question might be adequately answered using other than animal means, i.e., computer modeling, cell cultures, etc.**

2. **Animal Model and Species Justification:** It is important that you adequately justify that animals are necessary for attainment of the research/training objectives. Moreover, justify the selection of this particular animal model. Investigators should use the least sentient species that will permit the attainment of research objectives. Why was this particular animal chosen? Were there other animal models considered that are lower on the phylogenetic scale (e.g., mice instead of rabbits)? Is there a unique quality or usefulness about this species that warrants its selection for use?

3. **Laboratory Animals:** No response necessary to the title paragraph of this section.

a. **Genus & Species:**

b. **Strain/Stock:** If inbred or specialized animals are required, please use proper terminology.

c. Source/Vendor: Provide a preferred source for the animals. Procurement of animals from non-USDA licensed sources requires an exception to policy. Enter the source/vendors USDA license number if available.

d. Age:

e. Weight:

f. Sex:

g. Special Considerations: Specialized requirements for the research animals should be reflected here, i.e., SIV or herpes antibody free, Pasteurella free, etc.

h. Other:

4. Total Number of Animals Required:

(a)	mice	320
(b)	guinea pigs	175

All that is required in this section is the total number of animals to be used on the study. The number requested here should match exactly those described in para V. A., Experimental Design & General Procedures in the MATERIALS AND METHODS section. Keep in mind the number requested should be the minimum numbers necessary to complete the study, but must be sufficient to yield meaningful results. If too few animals are requested and statistical significance is not achieved, the animals will have been misused. Be certain to include animals necessary for controls or technique development, etc. If additional animals are needed due to technical or unavoidable circumstances, or to exploit a serendipitous finding, follow IACUC procedures for requesting approval for additional animals.

5. Refinement, Reduction, Replacement: The DoD is often required to provide specific examples of its alternatives initiatives. Does this protocol have any provisions that would qualify it to be identified as one that refines, reduces or replaces (3 R's) the use of animals? For example, does your study use statistical tests that require fewer animals, i.e., a modified LD50 test like Thompson & Weil, or are you using cell cultures, computer modeling or any other technique that will influence the numbers of animals required? Are you using animals lower on the phylogenetic scale? Please provide a short description of the features that you feel qualify the study as one that employs one of the "3 R's," or give a negative reply. No response is needed under the title paragraph of this section.

a. Refinement: The use of analgesia, or the use of remote telemetry to increase the quality and quantity of data

gathered or adjusted early endpoint for the animals are examples of refinements.

b. Reduction: Use of shared control groups, preliminary screening in non-animal systems or innovative statistical packages are examples of reductions.

c. Replacement: Non-animal systems that eliminate the use of animals are examples of replacement.

C. Technical Methods: These should be presented in sufficient detail, documented or referenced, so that the IACUC can adequately review the procedure and obtain a clear understanding of what is to be done, how the animal will be handled, and make a reasonable determination as to whether this proposed use of laboratory animals is in compliance with DoD regulations, guidelines, and federal law. No response is needed under the title paragraph of this section.

1. Pain: The law defines a painful procedure as one that would "reasonably be expected to cause more than slight or momentary pain or distress in a human being to which that procedure was applied, that is, pain in excess of that caused by injections or other minor procedures." **If a procedure involves pain or distress, the P.I. must consult with the attending veterinarian.** Respond N/A if the animals will experience "no pain or distress."

a. USDA (Form 18-3) Pain category:

This information is reported by the organization to the USDA on USDA Form VS 18-23. **The P.I. or primary user should estimate the number of animals that will be counted in each pain category.** There are many situations where there are animals in more than one category, i.e., control animals. If more than one species is requested in the proposal, reflect those animals in a duplicate table in this paragraph. **The total numbers reflected in these three categories should add up to the number and percent of animals requested for the entire protocol in para V.B.4.**

(1) No Pain _____(##)_____ % (Column C)

Studies involving no pain or distress beyond that expected on a momentary nature such as would occur with an injection, a deep palpation, grooming activities, etc.

(2) Alleviated Pain _____(##)_____ %
(Column D)

Procedures wherein anesthesia or analgesia will be administered to avoid or alleviate pain or distress. General anesthesia given for

surgical preparations, or the use of analgesia or anti-inflammatories would be examples for this category.

(3) Unalleviated Pain or Distress

_____ (#) _____ % (Column E)

Procedures where alleviation of pain or distress are contraindicated for some justifiable reason such as would confound the experimental results if drugs relieving pain were administered. Detailed justification for putting animals into this category is required below in para V.C.1.d.

b. Pain Alleviation: The attending veterinarian should be able to provide assistance in completing this section of the proposal.

(1) Anesthesia/Analgesia/Tranquilization: Describe the methods or strategies planned to alleviate pain or distress. If pain alleviation is planned, specify who will be administering the analgesics, anesthetics, or tranquilizers during the study. Provide agent, dosage, route & site, indication, needle size, etc.

(2) Paralytics: No use of paralytic agents without anesthesia is allowed unless scientifically justified by the P.I. and approved by the IACUC.

c. Alternatives to Painful Procedures:

(1) Source(s) Searched: e.g., AWIC, AGRICOLA, CAAT, MEDLINE, etc.

(2) Date of Search:

(3) Key Words of Search: e.g. Pain, surgery,

(4) Results of Search: Provide a narrative description of the results of the alternatives literature search. "Research facilities will be held responsible, if it is subsequently determined that an alternative to a painful procedure was available to accomplish the objectives of the proposed experiment." The Animal Welfare Act specifically states that the **"P.I. must provide a narrative description of the methods and sources, e.g., the Animal Welfare Information Center, MEDLINE, LIFE SCIENCES ABSTRACTS, AGRICOLA, AND BIOSIS that he\she used to determine that alternatives to the painful procedure were not available."** It is a requirement to perform the alternatives literature search and painful procedure justification even when animals are placed in the alleviated pain category (column D).

d. Painful Procedure Justification: Procedures causing more than transient or slight pain that are unalleviated, must be justified on a scientific basis in writing by the P.I. The pain must continue for only the necessary period of time dictated by the experiment, and then be alleviated, or the animal humanely euthanized. This paragraph must be completed if there are any animals listed in either the alleviated (column D) or the unalleviated pain or distress (column E) category in para V.C.1. **The P.I. must consult with the attending veterinarian or his or her designee in the planning of both alleviated and unalleviated painful procedures, and state it here.**

2. Prolonged Restraint: Describe and justify in detail any prolonged restraint (greater than twelve hours) intended for use during the study, e.g., primate chairs, restraint boards, metabolism cages, etc. Also describe habituation procedures for the prolonged restraint. This section is not intended for short-term actions such as rabbit restraint for bleeding, etc. If there is prolonged restraint involved, who will be restraining the animals, and for how long?

3. Surgery: Major operative procedures on non-rodent species, i.e., rabbits, monkeys, etc., should be conducted only in dedicated facilities intended for that purpose, and operated and maintained under aseptic conditions. Non-major operative procedures & all rodent surgery do not require a dedicated facility, but must be performed using aseptic technique, i.e., surgical gloves, mask, sterile instruments. A major operative procedure is one that "penetrates and exposes a body cavity, or causes permanent impairment of physical or physiological function." The animal care unit personnel should assist in defining the requirements of this portion of the law if necessary. No response required under the title paragraph of this section.

a. Procedure: Describe in detail any surgical procedures planned.

b. Pre- and Postoperative Provisions: Detail the provisions for both pre- and postoperative care, including provisions for post-surgical observations. Also include the provider of that care, and the location for the postoperative care.

c. Location: Give the location\room # for the proposed surgical procedure.

d. Multiple Survival Surgery Procedures: If multiple major operative procedures on the same animal are intended, they must be adequately justified for scientific reasons by the P.I. in writing.

(1) **Procedures:**

(2) **Scientific Justification:**

4. Animal Manipulations: Any injections, sampling procedures, or other manipulations of the animals necessary for the execution of the study must be described if not listed in section V. List needle sizes, routes of injection or withdrawal and anatomical location, e.g. 21 ga needle, SQ, IM, femoral vein, jugular vein etc., or the proposed method so that a reasonable evaluation of the appropriateness of the procedure can be made. You may furnish the committee a reference or SOP to document a particular procedure in lieu of a detailed description. You may wish to rearrange the subparagraphs of this section to suit your protocol. No response is needed under the title paragraph of this section.

a. Injections: There is no need to duplicate specific information already provided in section V.C.1.b., the Pain Alleviation, anesthesia/analgesia section of the proposal.

b. Biosamples: Cerebral taps, blood sampling, etc. List amounts taken and method for sampling. Procedures performed or biosamples obtained during a necropsy need not be described here.

c. Animal Identification: Microchip, tattoo, ear tags, cage cards, etc.

d. Behavioral Studies: Fully describe any intent to use aversive stimuli, food or water deprivation, etc, that would impact upon the animals in this study.

e. Other procedures: EKG's, radiology, aerosol exposure, etc.

5. Adjuvants: List any adjuvants and your plan for their use. Provide dosages & route.

6. Study Endpoint: What is the projected end point or termination of the study for the animals? Is death, euthanasia, or recovery expected; and what is the specific plan for determining when the animal experimentation phase will be stopped? You should ensure that unnecessary pain or distress is prevented by carefully considering "When is the experimental question answered?" so that the animals can be removed from the study as soon as feasible. Explain the plan for the disposition of surviving animals. **You must specifically address and justify any proposed use of death as an endpoint.**

7. Euthanasia: Explain the plan for euthanasia of the animals at the completion of the study and who will perform the procedure. The AWA defines euthanasia as "humane destruction of an animal by a method that produces rapid unconsciousness and subsequent

death without evidence of pain or distress, or a method that utilizes anesthesia produced by an agent that causes painless loss of consciousness and subsequent death." The current AVMA guidelines for euthanasia must be followed. Exceptions to the AVMA guidelines will be considered by the IACUC on a case-by-case basis. Exceptions must be scientifically justified by the P.I. in writing. The attending veterinarian will assist in selecting the best method for euthanasia if requested.

D. Veterinary Care: Attending veterinary care of lab animals receives particular emphasis in the AWA. The attending veterinarian of your facility will assist P.I.s with preparing this section if requested. No response is necessary to the title paragraph of this subsection.

1. Husbandry Considerations: The law specifically states that animal housing and living conditions must be appropriate to their species, and contribute to their health and comfort. Describe husbandry or refer to SOP. If known, list the location the animals will be routinely housed and the length of housing requirement. Personnel in the animal care unit should be able assist P.I.s in the preparation of the protocol sections dealing with animal care issues.

a. Study Room: If stay exceeds 12 hours.

b. Special Husbandry Provisions: Micro-isolators, metabolic cages, etc.

2. Attending Veterinary Care: Will the animals be observed daily or more frequently, and by whom? What is the plan if the animal becomes ill or debilitated during the study and requires supportive therapy? Will the animal be euthanized if it becomes critically ill or comatose, and by whom (study endpoint adjustment)? Justification for not providing supportive care for clinically ill animals is necessary.

3. Enrichment Strategy: Written justification for restricting enrichment programs or activity programs of dogs, cats, or nonhuman primates must be provided.

a. Dogs: Do you have any reason to restrict activity programs for dogs on this protocol that might be implemented by the animal care unit to comply with federal welfare regulations. If yes, justify.

b. Nonhuman Primates: Do you have any reason to prohibit environmental enrichment or enhancement strategies that might be implemented by the animal care unit to comply with federal welfare regulations. If yes, justify.

E. Data Analysis: List the statistical test(s) planned or the strategy intended to evaluate the data.

F. Investigator & Technician Qualifications/Training: List those animal procedures or manipulations described in the protocol that will be performed by each investigator or technician, and their training or qualifications to perform these procedures. Personnel conducting the "hands-on" animal procedures described in the protocol must be identified and appropriately trained and qualified to perform that procedure. **This is NOT questioning the P.I.'s PROFESSIONAL qualifications to conduct the research, but rather a requirement that personnel actually performing the research animal manipulations are technically competent, and thus are not inflicting unnecessary pain, distress, or injury to an experimental animal due to inexperience or improper technique.** Contact your attending veterinarian for assistance with this requirement.

VI. Biohazard/Safety: Provide a list of any potential biohazards associated with this proposal, e.g., viral agents, toxins, radioisotopes, oncogenic viruses, chemical carcinogens, etc. Explain any safety precautions or programs designed to protect personnel from biohazards, and any surveillance procedures in place to monitor potential exposures.

(Start new page here)

VII. ASSURANCES: The law specifically requires several written assurances from the P.I. It states that "research facilities will be held responsible if it is subsequently determined that an experiment is unnecessarily duplicative, and that a good faith review of available sources would have indicated as much."

(This section will state) As the Primary Investigator on this protocol I acknowledge my responsibilities and provide assurances for the following:

A. Animal Use: The animals authorized for use in this protocol will be used only in the activities and in the manner described herein, unless a deviation is specifically approved by the IACUC.

B. Duplication of Effort: I have made a reasonable, good faith effort to ensure that this protocol is not an unnecessary duplication of previous experiments.

C. Statistical Assurance: I assure that I have consulted with an individual who is qualified to evaluate the statistical design or strategy of this proposal, and that the "minimum number of animals needed for scientific validity are used."

D. Biohazard\Safety: I have taken into consideration, and I have made the proper coordinations regarding all applicable rules and regulations regarding radiation protection, biosafety, recombinant issues, etc., in the preparation of this protocol.

E. Training: I verify that the personnel performing the animal procedures/manipulations described in this protocol are technically competent and have been properly trained to ensure that no unnecessary pain or distress will be caused as a result of the procedures/manipulations.

F. Responsibility: I acknowledge the inherent moral and administrative obligations associated with the performance of this animal use protocol, and I assure that all individuals associated with this project will demonstrate a concern for the health, comfort, welfare, and well-being of the research animals. Additionally, I pledge to conduct this study in the spirit of the fourth "R" which the DoD has embraced, namely, "Responsibility" for implementing animal use alternatives where feasible, and conducting humane and lawful research.

(Signature Required)

(Primary Investigator)

G. Painful Procedures: (Include only if conducting research that will cause more than slight or momentary pain or distress (Column D or E by USDA classification) the following statement must follow.) **I am conducting biomedical experiments which may potentially cause more than momentary or slight pain or distress to animals that WILL BE relieved or WILL NOT (circle one) be relieved with the use of anesthetics, analgesics and/or tranquilizers.** I have considered alternatives to such procedures; however, using the methods and sources described in the protocol, I have determined that alternative procedures are not available to accomplish the objectives of the proposed experiment.

(Signature Required)

(Primary Investigator)

VIII. Enclosures: (Available for the attachment of the results of any literature searches, SOPs, references, or other documents pertinent to the protocol that you may wish to include. Local IACUC's should determine specific items to be included here.)

A. Literature Searches: DTIC, FEDRIP, MEDLINE, AGRICOLA, etc.

B. Pathology Addendum: Optional information

C. Pain Scoring Guidelines:

D. Adjuvant Policy:

PROTOCOL COVER SHEET

PROTOCOL TITLE:

PRINCIPAL INVESTIGATOR:

(Signature Required)

(Principal Investigator)

SCIENTIFIC REVIEW:

(Signature Required)

(Research Unit Chief/Directors signature)

ATTENDING/CONSULTING VETERINARIAN:

(Signature Required)

(Attending/Consulting Veterinarian)

STATISTICAL REVIEW: A person knowledgeable in statistics has reviewed the experimental design. (No response is required to the title paragraph of this section) (Inclusion of Signature Block is Recommended, but Optional)

(Statistician)

***OTHERS:** You may wish to add specific additional offices or signature blocks for individuals responsible for coordination or compliance issues pertinent to your facility or operation. (i.e. Co-investigators, Coordinating Departments, IACUC Chair, Biosafety Review etc.)

PROTOCOL TITLE:

PRINCIPAL INVESTIGATOR:

CO-INVESTIGATOR(S):

- I. **NON-TECHNICAL SYNOPSIS:**
- II. **BACKGROUND:**
 - A. **Background:**
 - B. **Literature Search:**
 - 1. **Literature Source(s) Searched:**
 - 2. **Date and Number of Search:**
 - 3. **Key Words of Search:**
 - 4. **Results of Search:**
- III. **OBJECTIVE\HYPOTHESIS:**
- IV. **MILITARY RELEVANCE:**
- V. **MATERIALS AND METHODS:**
 - A. **Experimental Design and General Procedures:**

- B. Laboratory Animals Required and Justification:
1. Non-animal Alternatives Considered:
 2. Animal Model and Species Justification:
 3. Laboratory Animals:
 - a. Genus & Species:
 - b. Strain/Stock:
 - c. Source/Vendor:
 - d. Age:
 - e. Weight:
 - f. Sex:
 - g. Special Considerations:
 - h. Other:
 4. Total Number of Animals Required:
 5. Refinement, Reduction, Replacement:
 - a. Refinement:
 - b. Reduction:
 - c. Replacement:
- C. Technical Methods:
1. Pain:
 - a. USDA (Form 18-3) Pain category:
 - (1) No Pain _____(##)_____ % (Column C)
 - (2) Alleviated Pain _____(##)_____ % (Column D)
 - (3) Unalleviated Pain or Distress
_____ (##) _____ % (Column E)
 - b. Pain Alleviation:
 - (1) Anesthesia/Analgesia/Tranquilization:
 - (2) Paralytics:
 - c. Alternatives to Painful Procedures:
 - (1) Source(s) Searched:
 - (2) Date of Search:
 - (3) Key Words of Search:
 - (4) Results of Search:
 - d. Painful Procedure Justification:
 2. Prolonged Restraint:
 3. Surgery:
 - a. Procedure:
 - b. Pre- and Postoperative Provisions:
 - c. Location:
 - d. Multiple Survival Surgery Procedures:
 - (1) Procedures:
 - (2) Scientific Justification:
 4. Animal Manipulations:
 - a. Injections:
 - b. Biosamples:
 - c. Animal Identification:
 - d. Behavioral Studies:
 - e. Other procedures:
 5. Adjuvants:
 6. Study Endpoint:

- 7. Euthanasia:
- D. Veterinary Care:
 - 1. Husbandry Considerations:
 - a. Study Room:
 - b. Special Husbandry Provisions:
 - 2. Attending Veterinary Care:
 - 3. Enrichment Strategy:
 - a. Dogs:
 - b. Nonhuman Primates:
- E. Data Analysis:
- F. Investigator & Technician Qualifications/Training:
- VI. Biohazard/Safety:

(Start new page here)

VII. ASSURANCES: As the Primary Investigator on this protocol I provide the following assurances:

A. Animal Use: The animals authorized for use in this protocol will be used only in the activities and in the manner described herein, unless a deviation is specifically approved by the IACUC.

B. Duplication of Effort: I have made a reasonable, good faith effort to ensure that this protocol is not an unnecessary duplication of previous experiments.

C. Statistical Assurance: I assure that I have consulted with an individual who is qualified to evaluate the statistical design or strategy of this proposal, and that the "minimum number of animals needed for scientific validity are used."

D. Biohazard\Safety: I have taken into consideration, and I have made the proper coordinations regarding all applicable rules and regulations regarding radiation protection, biosafety, recombinant issues, etc., in the preparation of this protocol.

E. Training: I verify that the personnel performing the animal procedures/manipulations described in this protocol are technically competent and have been properly trained to ensure that no unnecessary pain or distress will be caused as a result of the procedures/manipulations.

F. Responsibility: I acknowledge the inherent moral and administrative obligations associated with the performance of this animal use protocol, and I assure that all individuals associated with this project will demonstrate a concern for the health, comfort, welfare, and well-being of the research animals. Additionally, I pledge to conduct this study in the spirit of the fourth "R" which the DoD has embraced, namely, "Responsibility" for implementing animal use alternatives where feasible, and conducting humane and lawful research.

(Signature Required)

(Primary Investigator)

G. Painful Procedures: (Include above if conducting research that will cause more than slight or momentary pain or distress (Column D or E by USDA classification) the following statement must follow.) **I am conducting biomedical experiments which may potentially cause more than momentary or slight pain or distress to animals that WILL BE relieved or WILL NOT (circle one) be relieved with the use of anesthetics, analgesics and/or tranquilizers.** I have considered alternatives to such procedures; however, using the methods and sources described in the protocol, I have determined that alternative procedures are not available to accomplish the objectives of the proposed experiment.

(Signature Required)

(Primary Investigator)

VIII. Enclosures: (Available for the attachment of the results of any literature searches, SOPs, references, or other documents pertinent to the protocol that you may wish to include. Local IACUC's should determine specific items to be included here.)

A. Literature Searches: FEDRIP, DTIC, MEDLINE, AGRICOLA, etc.

B. Pathology Addendum: Optional information

C. Pain Scoring Guidelines:

D. Adjuvant Policy:

Appendix D

DoD Semiannual Program Review and Facility Inspection Checklist

DOD SEMIANNUAL PROGRAM REVIEW/FACILITY INSPECTION CHECKLIST-MANDATORY

Completion of this one-page checklist by the IACUC during the semi-annual program review and facility inspection is mandatory.

ORGANIZATION: _____ **DATE OF REVIEW:** _____

EVALUATION VIA CATEGORY	S	M	U	NA	EVALUATION VIA CATEGORY	S	M	U	NA
AAALAC History					Identification Records				
Administrative Commitment					Emergency, Weekend & Holiday Care				
Administrative Organization					Adequate Veterinary Care				
Institutional Policies					Preventive Medicine				
Animal Care & Use Committee					Animal Procurement				
Protocol Review Procedures					Quarantine Isolation				
Personnel Qualifications					Control of Animal Disease				
Personnel Hygiene					Diagnostic Resource				
Occupational Health Program					Anesthesia & Analgesia				
Animal Restraint					Surgery & Postsurgical Care				
Multiple Major Surgeries					Euthanasia				
Animal Husbandry					Physical Plan Arrangement/Cond.				
Housing/Caging & Pens					Support Areas				
Social Enrichment					Cage Sanitation Fac.				
Activity/Exercise					Storage Facilities				
Food/Water/Bedding					Surgery Facilities				
Sanitation					Animal Rooms				
Waste Disposal Methods					HVAC				
Vermin Control					Emergency Power				
Farm Facilities					Animal Use Laboratories				

KEY: S = Satisfactory; M = Minor Deficiency; U = Unsatisfactory/Major deficiency; NA = Not Applicable

USE OF CHECKLIST IN PROGRAM EVALUATION-- Completion of this one page checklist is mandatory. Any area that has minor or Major/Unsatisfactory deficiencies should be further explained on a separate page(s). Moreover, the listing of the minor or major deficiency should also include a plan of action for correction of the deficiency.

DETAILED OUTLINE OF CHECKLIST-- Utilization of this outline is optional. Attached is a detailed outline which follows this checklist. The outline includes most additional DoD requirements and is very similar to the program description outline used by organizations applying for AAALAC accreditation. This outline or one devised by your IACUC can be used to augment your semi-annual program reviews.

USE OF ROOM INSPECTION FORM-- Utilization of attached form is optional. The use of this form or one developed by your organization may be useful in augmenting your semi-annual program review.

MINORITY OPINIONS-- Utilization of attached form is optional. All minority opinions must be included in the IACUC report. In addition it is mandatory that a majority of IACUC members sign the semi-annual report.

There were / were not (circle one) minority opinions in this semi-annual review.

-OPTIONAL-

DETAILED OUTLINE OF CHECKLIST-- Utilization of this outline is optional. Attached is a detailed outline which follows this checklist. The outline includes most additional DoD requirements and is very similar to the program description outline used by organizations applying for AAALAC accreditation. This outline or one devised by your IACUC can be used to augment your semiannual program reviews.

A. General Comments AAALAC history, administrative commitment, administrative organization,

B. Institutional Policies

1. Monitoring the Care and Use of Animals

a. Institutional Animal Care and Use Committee

1) Composition- New DoD Directive states the minimum number for IACUC membership is 5. New DoD policy states requires those IACUCS with only one non-affiliated member the IACUC to also appoint an additional alternate non-affiliated member. New DoD policy states specific training requirements for non-affiliated IACUC members (8 hours).

2) Protocol review procedures- New DoD Directive and policies require use of DoD standard protocol format. New requirements include documentation of literature searches for DTIC, FEDRIP and other searches as required.

3) Review of programs for Care and Use of Animals- New DoD policy encourages Commanders/Directors/CEO's of DoD laboratories to invest in training at all levels for those that use animals.

b. USDA Report

2. Veterinary Care

a. Intensity -

b. Responsibilities of the Veterinarian(s) -

c. Involvement in monitoring the care of animals -

d. Involvement in monitoring use of animals -

3. Personnel Qualifications

a. Animal resource Professional/Management/ Supervisory Personnel -

b. Animal Care Personnel -

c. Research Staff -

d. Use of Hazardous Agents -

4. Personnel Hygiene

a. Work clothing provided -

b. Laundering of work clothing -

c. Shower and change facilities -

d. Eating, drinking, and smoking policies -

e. Eating, drinking, and smoking facilities -

5. Occupational Health and Safety Program

a. Content of program -

b. Program oversight -

c. Participation by staff -

d. Training on zoonosis and personal hygiene -

6. Experimentation involving Hazardous Agents

7. Animal Restraint -

8. Multiple Major Surgical Procedures -

C. Laboratory Animal Husbandry

1. Housing

a. Caging and pens -

- b. Social enrichment -
- c. Activity/exercise -
- d. Micro- & Macroenvironments -

2. Food

- a. Type -
- b. Vendor quality control -
- c. Storage -
- d. Type of feeders -
- e. Institutional quality control -

3. Bedding

- a. Type -
- b. Appropriateness for how used -
- c. Storage facilities -
- d. Quality control -

4. Water

- a. Source - Satisfactory.
- b. Treatment - Satisfactory.
- c. Quality control procedures -

5. Sanitation

- a. Cage & pan litter changing -
- b. Portable cage sanitation
 - 1) Frequency -
 - 2) Procedures and agents -
 - 3) Monitoring and effectiveness -
- c. Pens, Stalls, etc. -
- d. Sanitation of feeding implements -
- e. Watering Implements
 - 1) Water Bottles -
 - 2) Automatic watering system -
- f. Sanitation of transport cages and vehicles -
- g. Room sanitation -
- h. Waste disposal methods -
- i. Vermin control -

6. Animal Identification

- a. Methods for identification of each species -
- b. Information of cage cards -
- c. Individual animal records -

7. Provisions for Emergency, Weekend and Holiday Care

- a. Qualifications of individuals providing care -
- b. Procedures performed -
- c. Monitoring of environmental systems -

D. Veterinary Care

1. Preventive Medicine

- a. Animal procurement -
- b. Quarantine, Stabilization and Isolation -
 - 1) Receiving and initial evaluation procedures -
 - 2) Quarantine facilities
 - a) For random source animals -
 - b) For purpose bred animals -

- 3) Quarantine procedures -
- c. Separation by species, source and health status -
- 2. Surveillance, Diagnosis, Treatment, and Control of Animal Disease**
 - a. Program
 - 1) Daily observation of animals -
 - 2) Procedures for providing veterinary care -
 - 3) Medical Records maintenance procedures -
 - 4) Preventive medicine program for each species -
 - 5) Animal Health monitoring -
 - b. Diagnostic Resources
 - 1) Clinical Laboratory -
 - 2) Necropsy/histology -
 - 3) Radiology -
 - 4) Use of available diagnostic resources including commercial laboratories -
- 3. Anesthesia and Analgesia**
 - a. Agents used for each species -
 - b. Guidelines provided by the Veterinarian -
 - c. Monitoring the use of A & A -
 - d. Training and experience of personnel who perform anesthesia -
 - e. Safety procedures for use of explosive/flammable agents -
 - f. Waste anesthetic gas scavenging -
- 4. Survival Surgery and Postsurgical Care**
 - a. Non-rodent mammalian species
 - 1) Professional supervision -
 - 2) Qualifications of persons performing the surgery -
 - 3) Qualifications of surgical technicians -
 - 4) Aseptic Techniques -
 - 5) Postoperative care -
 - 6) Maintenance of PO care records -
 - b. Rodent species - use of cap, mask, surgical scrub, sterilized instruments used, hair clipped, .
 - c. Non-survival surgeries -

E. Physical Plant

- 1. Overview of General Arrangement and Condition of Facility**
- 2. Support Areas**
 - a. Clean cage storage -
 - b. Storage Areas -
 - c. Waste disposal facilities -
 - d. Lounge area for animal care personnel -
 - e. Administrative space -
 - f. Cage sanitation facilities -
 - 1) Interior surfaces -
 - 2) Sanitation equipment -
 - 3) Environmental conditions for personnel -
 - g. Surgery facilities
 - 1) Areas for
 - a) Surgery -
 - b) Animal preparation -
 - c) Dressing rooms -
 - d) Surgeon preparation -
 - e) Postoperative care -

DoD Semiannual Program Review/Facility Inspection

3. Animal Rooms

- a. Interior surfaces -
- b. Lighting - Satisfactory.
- c. HVAC -

4. Other Features

- a. Emergency power -
- b. Environmental monitoring
 - 1) Animal rooms air flow -
 - 2) Relative air pressures -
 - 3) Temperature -
 - 4) Humidity -
- c. Security -

5. Miscellaneous Animal Care and Use Equipment

F. Special Considerations

- 1. Genetics and Nomenclature -
- 2. Facilities and Procedures for Animal Research Involving Hazardous Agents -
- 3. Farm Animals -

G. Study Areas Visited -

H. Laboratories Visited -

-OPTIONAL-
USE OF ROOM INSPECTION FORM--Utilization of attached form is optional. The use of this form or one developed by your organization may be useful in augmenting your semi-annual program review.

Building _____

=====

ROOM _____	Animal Holding Area	Lab	Other
-------------------	---------------------	-----	-------

=====

ROOM _____	Animal Holding Area	Lab	Other
-------------------	---------------------	-----	-------

=====

ROOM _____	Animal Holding Area	Lab	Other
-------------------	---------------------	-----	-------

=====

ROOM _____	Animal Holding Area	Lab	Other
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=====

GENERAL COMMENTS:

There were / were not (circle one) minority opinions in this semi-annual review.

The Animal Welfare Act requires IACUCs to review and inspect laboratory animal care and use programs on a semiannual basis. This form facilitates compliance with the requirement that at least a majority of members of the IACUC sign the semiannual report, and have a opportunity to express a minority opinion to the report. Minority opinions should be appended to the report in writing.

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Appendix E

U.S. Government Principles for Animal Use

Appendix E

U.S. Government Principles for Animal Use

Interagency Research Animal Committee's

U.S. Government Principles for the Utilization and Care of Vertebrate Animals Used in Testing, Research and Training

The development of knowledge necessary for the improvement of the health and well-being of humans as well as other animals requires in vivo experimentation with a wide variety of animal species. Whenever U.S. Government agencies develop requirements for testing, research, or training procedures involving the use of vertebrate animals, the following principles shall be considered; and whenever these agencies actually perform or sponsor such procedures, the responsible institutional official shall ensure that these principles are adhered to:

- I. The transportation, care and use of animals should be in accordance with the Animal Welfare Act (7 U.S.C. 2131 et. seq.) and other applicable Federal laws, guidelines, and policies.¹
- II. Procedures involving animals should be designed and performed with due consideration of their relevance to human or animal health, the advancement of knowledge, or the good of society.
- III. The animals selected for a procedure should be of an appropriate species and quality and the minimum number required to obtain valid results. Methods such as mathematical models, computer simulation, and in vitro biological systems should be considered.
- IV. Proper use of animals, including the avoidance or minimization of discomfort, distress, and pain when consistent with sound scientific practices, is imperative. Unless the contrary is established, investigators should consider that procedures that cause pain or distress in human beings may cause pain or distress in other animals.
- V. Procedures with animals that may cause more than momentary or slight pain or distress should be performed with appropriate sedation, analgesia, or anesthesia. Surgical or other painful procedures should not be performed on unanesthetized animals paralyzed by chemical agents.
- VI. Animals that would otherwise suffer severe or chronic pain or distress that cannot be relieved should be painlessly killed at the end of the procedure or, if appropriate, during the procedure.
- VII. The living conditions of animals should be appropriate for their species and contribute to their health and comfort. Normally, the housing, feeding, and care of all animals used for biomedical purposes must be directed by a veterinarian or other scientist trained and experienced in the proper care, handling, and use of the species being maintained or studies. In any case, veterinary care shall be provided as indicated.
- VIII. Investigators and other personnel shall be appropriately qualified and experienced for conducting procedures on living animals. Adequate arrangements shall be made for their inservice training, including the proper and humane care and use of laboratory animals.
- IX. Where exceptions are required in relation to the provisions of these Principles, the decisions should not rest with the investigators directly concerned but should be made, with due regard to Principle II, by an appropriate review group such as an institutional animal research committee. Such exceptions should not be made solely for the purposes of teaching or demonstration.

¹ For guidance throughout these Principles the reader is referred to the Guide for the Care and Use of Laboratory Animals prepared by the Institute of Laboratory Animal Resources, National Research Council.

Published in the Federal
Register, May 20, 1985,
Vol. 50, No. 97, by the
Office of Science and
Technology Policy

Appendix F

Benefits of DoD Animal Care and Use Programs

Appendix F

Benefits of DoD Animal Care and Use Programs

I. MEDICAL

Infectious Diseases

Determination of the ability of Old World sand flies to transmit New World Leishmania
Development of safe and effective liposomes as carriers of drugs and vaccines
Provision of antigens for assays for dengue and Japanese encephalitis
Completion of neutralizing antibody studies with regard to dengue viral research
Conduct of preclinical testing of various candidate vaccines
Study of immune response to Shigella antigen preparations
Definition of optimal conditions for lipopolysaccharide detoxification and immunization scheduling
Development of recombinant dengue vaccines toward generating safe and effective vaccine candidates
Development of methods to screen and monitor dengue infection for eventual vaccine testing
Development of a ground-breaking method for culturing an important malaria pathogen
Determination of prevalence of the sand fly and other pathogens in U.S. troops stationed overseas
Determined schistosomiasis antigens to be found in saliva, a diagnostic breakthrough
Observation of, for vaccine safety testing, the long-term T-cell immune response to vaccination
Isolation and characterization of hantavirus recovered from feral rodents
Identification of meningitis/encephalitis disease associated with Rift Valley fever in Egypt
Identification of the most active fraction(s) in mosquito components inducing host resistance
Development of DNA vaccines, anticipated to be a new and highly cost effective method of vaccination
Development of a model for human calciviral infection
Establishment of mosquito breeding colonies for the study of malaria and for diagnostic purposes
Study of mycoplasmas and their possible involvement in toxic shock syndrome
Study of blood cell proliferation growth factors toward controlling blood cell regeneration
Demonstrated that intranasal immunization can be superior to either oral or gastric immunization
Performed preclinical vaccine safety and efficacy testing

Medical Chemical Defense

Development of a computer model to predict agent toxicokinetics regardless of the animal species used
Identification of mechanisms for, and countermeasures to, blister agent intoxication
Design, expression and production of enzymes that can scavenge nerve agents in blood
Development and use of software for physiologically based pharmacokinetic models of soman nerve agent stereoisomer distribution and inhalation exposure
Development of safer, more effective antiseizure and anticonvulsant compounds
Determined that clinically employed antimalarial drugs can reduce the rate of botulinum toxin paralysis
Development of sensitive assays for determining the actions of botulinum toxin on neurotransmitter release
Conducted preclinical drug tests to explore drug metabolism, detection, and disposition
Demonstrated that exogenous acetylcholinesterase nerve agent scavenger produces little behavioral impact on a learning task
Development of a prototype system to monitor cyanide exposure effects on hemoglobin

Medical Biological Defense

Development of monoclonal antibodies that can protect animals from the lethal effects of low levels of three serotypes of botulinum toxin
Development of botulinum toxin vaccine using a mouse model
Demonstration of the effectiveness of a new vaccine candidate for inhaled plague bacteria
Development and characterization of surrogate markers of protection against anthrax
Characterization of the immunogenicity of cholera toxin
Demonstrated that a single dose of a promising antiviral drug protects against monkeypox infection
Identification of two cytokines that are important to *Staphylococcus B* infection
Identification of the most useful current antibiotics for treatment of glanders disease in humans

Human Systems Technology

Provided evidence that four different compounds have neuroprotective properties on ischemic retina
Demonstrated heat-shock treatment to differentially protect cultures from different brain regions
Performed research to understand the physiological mechanisms underlying circulatory shock
Tested pharmaceutical agents for their prevention or reversal behavioral changes associated with conditioned defeat
Provided further evidence that tyrosine can alleviate negative effects of acute stressors (i.e., hyperthermia) on performance

Combat Casualty Care

Trained physicians in combat casualty care
Evaluated the efficacy and mechanisms of action of several distant and novel neuroprotective agents
Provided critical training to combat medical personnel involved in overseas military operations
Development of novel pharmaceutical approaches to the treatment of brain injury

Ionizing Radiation

Developed enhanced treatments for radiation-associated infections using immune system stimulators
Developed new strategies for preventive treatment of both acute and chronic radiation injuries based on mechanisms of cellular and molecular injury
Initiated studies to assess the cancer-causing potential and immune system effects of depleted uranium
Determined that ionizing radiation induces a specific deletion in mitochondrial DNA
Evaluated a new delivery platform for cytogenetic-based radiation dose assessment in individuals
Determined that the nerve agent prophylactic pyridostigmine and sub-lethal radiation synergistically cause a redistribution of blood flow within the body

Other Medical Research

Determination, at military installations, of the presence of rodent-borne viral pathogens
Conducted EPA-mandated studies of Trichloromelamine, Pouch A, Food Service Disinfectant
Development of a guinea pig model of underwater HELF sound exposure with which to study effects on vestibular function

II. CLINICAL INVESTIGATIONS

Clinical Medicine

Demonstrated the effect of hypertension on kidney function and hormonal response
Determination of strategies for the treatment of inner ear vestibular effects from Meniere's disease, and damage from sound-induced trauma
Determination of how certain hormones may affect bone resorption
Determination of the efficacy of drug countermeasures against brown recluse spider bites
Study of the protective effects of estrogen and its role in the development of atherosclerosis
Development of a model of plaque rupture and thrombosis

Demonstrated efficiency of algal fatty acid compounds against methicillin-resistant *Staphylococcus aureus*
Developed a new assay to detect microbial diseases which was used to characterize diseases in wild rats on Kwajalein Island
Studied the effects of residual calcium in commonly used bone grafts toward optimizing bone growth
Determined efficacy of the selectin-inhibiting drugs, to prevent or reduce renal ischemia-reperfusion injury from trauma and/or organ transplantation

Clinical Surgery

Study of orthopedic protocol regarding optimal postoperative care following achilles tendon repair
Demonstrated that CO₂ laser resurfacing performed concurrently with skin-flap elevation adversely impacts skin flap survival
Conducted pilot study toward the development of bioabsorbable pins
Determined that transthoracic defibrillation can be effectively carried out using some dermal replacement systems, and determined the optimal system in high-risk burn patients
Studied bone regeneration toward developing treatment of bone trauma or disease, or periodontal disease
Used a tibial bone model to simulate placement of dental implants in the human mandible
Performed studies to improve methods for the repair of intestinal wounds
Development of a stenting method for keeping a lower limb perfused after a severe traumatic injury
Demonstrated the benefit in using a combined drug and perfusion pump augmentation in decreasing the incidence of post-operative kidney failure in a surgical procedure for aortic aneurysms
Evaluation of the efficacy of a new material proposed for use in treating injuries to the knee cartilage
Evaluation of drainage tube position on the completeness of fluid drainage from the thoracic cavity
Obtained promising preliminary results using autologous platelet gel as a biological sealant in lung surgery
Conducted preclinical research to increase quality of life for millions of glaucoma patients
Investigation of different types of stents, stent coatings, or irradiated stents to reduce incidence of restenosis
Determination of the efficacy of cyanoacrylate adhesive over the standard suture technique in closing stellate scleral lacerations

Other Clinical Investigations

Training in microvascular surgical techniques required in performance of gynecological procedures
Instruction of Advanced Trauma Life Support Course to physicians
Training of physicians in vessel and nerve resection techniques
Training of medical corpsmen

III. NON-MEDICAL

Physical Protection and Detection

Determining the efficacy of new technology to protect aircrew members from laser eye injury
Determining the cancer-causing potential of a commonly-used ultrawide radiowave radiation band source
Developed several biological detectors using monoclonal and polyclonal antibodies

Other Non-Medical RDT&E

Identification of a class of compounds that promote wakefulness without producing subsequent hypersomnolence
Determining the hypothetical role of sleep in neuroprotection
Ensuring that the products of chemical agent destruction will present no health hazards
Determining mechanisms by which the circadian clock operates at the cellular level to prevent sleepiness

Identification of molecules that regulate sleep so as to prevent sleep disturbances
Studied the impacts of dermal exposure to jet fuel on immune response
Determination of baseline electrophysiological data in selected animal species to standardize methods and determination of the limitations on anesthetics for each animal
Studied hibernating mammals to identify molecules that regulate metabolism, body temperature, and sleep
Development and testing of transplantation model to study the therapeutic potential of neural stem cells to repair central nervous system damage after traumatic injury
Identification of genetic factors that determine circadian behavior
Employing automated fish biomonitoring system to monitor remediation operations at contaminated sites

IV. TRAINING AND INSTRUCTIONAL

Training, Education, and/or Instruction for Personnel

Training to ensure quick and safe intubation of newborn infants
Training of surgical residents in microvascular, microsurgical procedures used in trauma care
Training in Advanced Trauma Life Support to prepare enlisted combat medics for battlefield injury care
Training of medical/surgical residents and pediatric nurses in neonatal resuscitation and care
Training of military medical residents and other health care professionals trained in a variety of procedures such as tracheotomy, splenectomy, hemorrhage control, fracture repair, intestinal anastomosis, etc.
Training of military veterinarians and veterinary technicians trained in emergency procedures critical to the provision of care to military working dogs
Training in pediatric bronchoesophagology on retrieving foreign bodies from compromised airways
Training in open and laparoscopic surgical techniques

Appendix G

**Journals and Proceedings with DoD Animal Research
Publications by Research Category**

Appendix G

Journals and Proceedings with DoD Animal Research Publications by Research Category

JOURNALS

MEDICAL

Infectious Disease

Annals of Tropical Medicine Parasitology
Archives of Virology
Clinical and Diagnostic Laboratory
Immunology
Clinical Infectious Diseases
Experimental Cell Research
Human Gene Therapy
Infection and Immunity
Infectious Immunology
Journal of Clinical Investigation
Journal of Experimental Medicine
Journal of General Virology
Journal of Immunology
Journal of Infectious Immunology
Journal of Medical Entomology
Journal of the American Mosquito Control
Association
Journal of the American Veterinary Medical
Association
Journal of Virology
Pediatric Research
The American Journal of Tropical Medicine
and Hygiene
Ultrastructural Pathology

Medical Chemical Defense

Archives of Toxicology
Brain Research Bulletin
Drug Metabolism and Disposition
Epilepsy Research
Experimental Lung Research
In Vitro and Molecular Toxicology
Molecular and Chemical Neuropathology
Neurotoxicology
Pharmacology, Biochemistry, and Behavior
Skin Research and Technology
Toxicology Methods

Medical Biological Defense

American Journal of Tropical Medicine and
Hygiene
Antimicrobial Agents and Chemotherapy
Cell Vision
Contemporary Topics in Laboratory Animal
Science

Hybridoma
Infection and Immunity
International Immunology
Journal of Biochemistry
Journal of Clinical Microbiology
Journal of Immunology
Journal of Infectious Disease
Microbial Pathogen
Nature
Pathophysiology
Pharmacology, Biochemistry, and Behavior
Plasmid
Vaccine
Veterinary Pathology
Virology

Military Operational Medicine

American Chemical Society
American Journal of Physiology
Biochemical and Biophysical Research
Communications
Biology of the Cell
Brain Research
Chronobiology International
FASEB Journal
Journal of Neurophysiology
Journal of Applied Physiology
Journal of Biological Rhythms
Journal of Immunology
Physiology and Behavior
Toxicological Application in Pharmacology
Undersea and Hyperbaric Medicine
Journal of Thermal Biology

Combat Casualty Care

Acute Respiratory Distress Syndrome:
Cellular and Molecular Mechanisms and
Clinical Management
American Journal of Physiology
American Journal of Tropical Medicine and
Hygiene
Archives of Surgery
Biochemical and Biophysical Research
Communications
Biophysics Journal
Critical Care Medicine

FASEB Journal
 Graft
 Journal of Immunology
 Journal of Laboratory Clinical Medicine
 Journal of Leukocyte Biology Supplement
 Journal of Molecular and Cellular
 Cardiology
 Journal of Neuroimmunology
 Journal of The American Medical
 Association
 Journal of Trauma
 Molecular Brain Research
 Nature Medicine
 Pulmonary Edema
 Shock
 Vascular Endothelium: Pharmacologic and
 Genetic Manipulation

Ionizing Radiation

Environmental Health Perspectives
 Journal of Radiation Research
 Radiation Research

OTHER MEDICAL RDT&E

Medical Free Electron Laser

American Journal of Ophthalmology
 Annals of Surgery
 Experimental Eye Research
 Journal of Investigative Dermatology
 Journal of Surgical Research
 Lasers in Surgery and Medicine
 Photochemistry and Photobiology

CLINICAL INVESTIGATIONS

Clinical Medicine

American Journal of Physiology
 American Journal of Respiratory and
 Critical Care Medicine
 American Journal of Rhinology
 Brain Research
 Cell Growth Differences
 Circulation
 European Journal of Pharmacology
 FASEB Journal
 Inflammation
 Journal of American College of Cardiology
 Journal of Biological Chemistry
 Journal of Cancer Resistance
 Journal of Comprehensive Neurology
 Journal of Immunology
 Journal of Inflammation

Journal of Investigative Medicine
 Journal of Neuroscience
 Journal of Neuroscience Methods
 Journal of Nuclear Medicine
 Journal of Pharmacological Experimental
 Therapy
 Journal of the American Society of Nephrologists
 Journal of Trauma
 Military Medicine
 Microcirculation
 Otolaryngology Clinic of North America
 Otolaryngology-Head and Neck Surgery
 Pediatric Research
 Pharmacological Biochemistry Behavior
 Procedural National Academy of Science, USA
 Science

Clinical Surgery

American Journal of Surgery
 Circulation
 Clinical Chemistry
 Digestive Surgery
 Journal of the U.S. Army Medical Department
 Journal of Trauma
 Laryngoscope

Training, Education, and/or Instruction for Personnel

Cardiovascular Catheterization Diagnostics

Alternatives

Marine Mammal Science

NON-MEDICAL

Physical Protection

Annual Review of Neuroscience
 Archives of Ophthalmology
 Bioelectromagnetics
 Biosensors and Bioelectronics
 Brain and Language
 International Journal of Radiation Biology
 Investigative Ophthalmology and Visual Science
 Journal of Applied Physics
 Journal of Applied Physiology
 Journal of Biomechanics
 Journal of Comparative Physiology
 Journal of Laser Applications
 Journal of Neurophysiology
 Nature Neuroscience
 Progress in Quantum Electronics
 Radiation Research
 Science

OTHER ANIMAL USE CATEGORIES

Jet Lag and Wakefulness

Brain Research

Cell

European Journal of Pharmacology

Journal of Biological Rhythms

Journal of Neuroscience

Journal of Pharmacological and
Experimental Therapeutics

Neuroscience

Solvent and Metal Toxicity

American Journal of Physiology: Lung

Cellular and Molecular Physiology

Free Radicals in Biology and Medicine

In Vitro Molecular Toxicology

Jet Fuel Toxicity

Inhalation Toxicology

Marine Biology

Journal of Experimental Biology

Toxicology

Drug and Chemical Toxicology

Journal of Toxicology and Environmental
Health

Neurotoxicology

The Toxicologist

Toxicological Methods

Toxicological Science

Toxicology and Applied Pharmacology

Physiology

Journal of Neuroscience

Sleep

Environmental

Environmental Toxicology and Chemistry

Biorobotics

American Zoologist

Journal of Experimental Biology

Undersea Medicine

Journal of Biological Chemistry

Journal of Neuroscience

PROCEEDINGS

MEDICAL

Infectious Disease

97th Annual Meeting of the American
Society of Tropical Medicine and
Hygiene

98 Schistosomiasis Research Project,
International Conference

Proceedings of the National Academy of
Sciences

Military Operational Medicine

Experimental Biology Meeting San

Francisco, CA, April 1998

Society for Neuroscience 1998

Combat Casualty Care

Proceeding of the National Academy of
Sciences

Ionizing Radiation

21st Army Science Conference

Proceedings of the 1996 ERDEC Scientific
Conference on Chemical and Biological
Defense Research

Proceedings of the 1996 Workshop on
Assessment, Prophylaxis, and Treatment

CLINICAL INVESTIGATIONS

Clinical Medicine

American Academy of Periodontology
District 8 Annual Meeting

Proceedings of the National Academy of
Sciences

Clinical Surgery

21st Army Science Conference

38th International Conference on

Antimicrobial Agents and

Chemotherapy (ICAAC)

Academy of Periodontology District 8
Annual Meeting

First International Conference on Chemistry
of Antibiotics and Related Microbial
Products

Alternatives

International Association for Aquatic
Animal Medicine Proceedings

Proceedings of SPIE (Detection and
remediation technologies for mines and
minelike targets II)
Proceedings Society of Toxicology
The World Marine Mammal Science
Conference

NON-MEDICAL

Physiology
Proceedings of the National Academy of
Science

Appendix H

**DoD Inspector General Recommendations on
the Use of Animals in DoD Medical Research Facilities
and Contract Research Facilities
February 1994**

Appendix H

DoD Inspector General Recommendations on the Use of Animals in DoD Medical Research Facilities and Contract Research Facilities

MEDICAL RESEARCH FACILITIES

Recommendation 1: The Director of Defense for Research and Engineering, in coordination with the Assistant Secretary of Defense (Health Affairs), should issue Department of Defense policy that requires every Department of Defense research facility to:

1. Support, and as necessary develop, animal care and use training programs, and encourage certification for all personnel involved in the care, use, and treatment of the animals; and
2. Develop a formal checklist to be used by the Institutional Animal Care and Use Committee when conducting its semiannual inspection. The published reports should document use of the checklist. All members of the Institutional Animal Care and Use Committee should sign the report that also includes a statement indicating there are or are not minority opinions.

Recommendation 2: The Director of Defense for Research and Engineering, in coordination with the Assistant Secretary of Defense (Health Affairs) and the General Counsel, Department of Defense, should provide clear Department of Defense guidance concerning the requirements and qualifications of the non-affiliated member of the Institutional Animal Care and Use Committee. The guidance should establish eligibility requirements, professional qualification, and characteristics for committee members, and set the minimum number of non-affiliated members desired.

Recommendation 3: The Director of Defense for Research and Engineering, in coordination with the Assistant Secretary of Defense (Health Affairs), should direct the Armed Services Biomedical Research Evaluation and Management Committee to develop a standardized, comprehensive Department of Defense research protocol request form and require its use by all Department of Defense research facilities.

Recommendation 4: The Director of Defense for Research and Engineering, in coordination with the Assistant Secretary of Defense (Health Affairs), should ensure each research facility commander is provided with information concerning the commendable practices identified by the inspection teams for consideration in their animal care and use program.

CONTRACT RESEARCH FACILITIES

Recommendation 1: The Director of Defense Research and Engineering, in coordination with the Assistant Secretary of Defense (Health Affairs), should issue Department of Defense policy that requires the Military Departments and the research facilities operated by the Office of the Secretary of Defense to complete the following tasks before awarding any contract or grant that involves research using any live animals:

1. All extramural research proposals using live animals should be reviewed by a veterinarian trained and knowledgeable about laboratory animal medicine to ensure compliance with all Federal laws, and Department of Defense regulations and guidelines concerning the care and use of animals.

2. To ensure the facility is complying with the requirements in the Animal Welfare Regulation, the Department of Defense funding agency should contact the United States Department of Agriculture to obtain copies of the most recent inspection reports for a facility under consideration for a contract or grant.

Recommendation 2: The Director of Defense Research and Engineering, in coordination with the Assistant Secretary of Defense (Health Affairs), should issue Department of Defense policy that requires the Military Departments and research facilities operated by the Office of the Secretary of Defense to perform the following tasks after a contract or grant that involves live animal use is awarded:

1. A veterinarian knowledgeable about laboratory animal medicine should conduct site visits to evaluate the animal care and use program at contract research facilities using non-human primates, marine mammals, dogs, or cats; conducting research deemed sensitive; or cited by the United States Department of Agriculture as a research facility under investigation. The policy should include the requirements for the initial site visit and the conditions for follow-on site visits.
2. To ensure continued compliance with the Animal Welfare Regulation, the Department of Defense funding agency should contact the United States Department of Agriculture on a routine basis to obtain a copy of the most recent annual inspection report for each facility with an active contract.

Recommendation 3: The Director of Defense Research and Engineering, in coordination with the Assistant Secretary of Defense (Health Affairs), should direct the Military Departments and the research facilities operated by the Office of the Secretary of Defense to require that all contractor proposals for research using live animals include all the information contained in the standardized Department of Defense protocol request format.

Appendix I

Occupations/Avocations of Nonaffiliated IACUC Members

Appendix I

Occupations/Avocations of Nonaffiliated IACUC Members

<u>Occupation/Avocation</u>	<u>No. of Times Represented</u>
Accountant	2
Administrator	7
Animal Science Engineer/Technician	3
Attorney	3
Biologist	5
Biostatistician/Statistician	6
Chaplain/Clergy	8
Chemist	1
Contracting Specialist	1
Dance/Exercise Instructor	1
Game Warden	2
Geologist	1
Historian	1
Homemaker	1
Instructor/Teacher	4
Interior Designer	1
Librarian	1
Medical Technician	1
Military	3
Nurse	2
Personnel Manager	2
Physician	2
Psychologist	2
Purchasing Agent	1
Safety Engineer	1
Salesman	1
Supply Manager	1
Veterinarian	15
Wildlife Technician	3

Appendix J

Dissemination of Information on Animal Care and Use

Appendix J

Dissemination of Information on Animal Care and Use

- Posters throughout the facility advising employees and the public on procedures for filing animal care and use complaints emphasizing that individuals do not have to use the chain of command but can go directly to the Institutional Animal Care and Use Committee (IACUC) chairman or the Inspector General (IG)
- Annual briefings to all facility personnel on the IG complaint process
- Notices posted on bulletin boards throughout the facility on how to register a complaint
- Mandatory investigator training courses
- Mandatory monthly seminars
- Researchers and technicians required to have documented appropriate training before performing procedures on animals
- Requirement for research staff and graduate students to complete training courses on the humane and ethical use of animals prior to engaging in research activities
- Provision of operating instructions and manuals to each investigator
- Posters announcing availability of anonymous “hot line” for registering concerns/complaints
- Provision of library resources, including books, manuals, and videotapes
- Provision of regulatory and policy documents
- Provision of journal and newsletter subscriptions
- Provision of investigators’ procedural handbooks
- Briefings and veterinarian-directed discussions at IACUC meetings
- Provision of orientation training for new IACUC members

Examples of subjects listed in the context of training topics reported in FY98:

- | | |
|--|--|
| * Alternatives to animal use | * Minimizing animal use |
| * Alternatives to death as an endpoint | * Occupational health and safety |
| * Animal husbandry | * Prevention and control of zoonosis |
| * Animal models | * Procedural techniques |
| * Distress minimization | * Proper use of analgesics |
| * Ethics and animal welfare | * Proper use of anesthetics |
| * Euthanasia | * Proper use of tranquilizers |
| * Handling and restraint | * Protocol training |
| * Informational services and resources | * Reporting of deficiencies in animal care and use |
| * Legal and policy issues pertinent to animal care and use | * Statistical analysis |
| | * Surgical techniques |

Appendix K

The 1998 WRAIR DoD Laboratory Animal Workshop Schedule

Appendix K

The 1998 WRAIR DoD Laboratory Animal Workshop Schedule

Issues in Laboratory Animal Care and Use Workshop

General Information – The Issues in Laboratory Animal Care and Use Workshop provides an overview of the WRAIR's Laboratory Animal program. The course focuses on ethical, regulatory, and humane considerations for writing and implementing animal use protocols, and will examine public and scientific concerns surrounding the use of animals in research. There is no laboratory portion associated with this workshop; however, it does include an optional 20-minute tour of the animal facility. The course is open to investigators, technicians, and administrative personnel. Class size is limited to 20.

Time: 0830-1100

Schedule:

30 January 1998

26 June 1998

1 October 1998

29 January 1999

Writing an Animal Use Protocol Using the DoD Template Workshop

General Information – Effective 1 October 1995, all DoD research institutes were required to utilize a standard protocol template when writing animal use protocols. This workshop will review those requirements and the protocol review process. The course is open to investigators, technicians, and administrative personnel. Participants are invited to bring their lunches to the workshop.

Time: 1200-1300

Schedule:

30 January 1998

26 June 1998

1 October 1998

29 January 1999

Nonhuman Primates & Safety Badge Class

General Information – The Nonhuman Primates & Safety Badge Class includes a didactic, safety, and laboratory portion, focusing primarily on macaques. Individuals will take a short exam covering the special safety measures required for work with nonhuman primates. Upon passage of the exam, individuals will use a nonhuman primate room entrance authorization/medical alert badge. Further information about nonhuman primate safety issues or training in New World and other Nonhuman Primate species can be obtained by contacting the Division of Veterinary Medicine at (301) 295-7016.

Time: 0830-1230

Schedule:

16 January 1998

20 March 1998

14 May 1998

16 July 1998

2 October 1998

3 December 1998

Rodent Class (Rats, Mice, Guinea Pigs)

General Information – The Rodent Class is a general species specific course that includes both a didactic and laboratory portion.

Time: 0830-1300

Schedule:

2 April 1998

8 May 1998

12 June 1998

2 July 1998

6 August 1998

3 September 1998

6 November 1998

11 December 1998

Lagomorph Class

General Information – The Lagomorph Class is a general species specific course that includes both a didactic and laboratory portion.

Time: 0830-1300

Schedule:

26 February 1998

9 April 1998

19 June 1998

17 July 1998

11 September 1998

13 November 1998

15 January 1999

Ovine Class

General Information – The Ovine Class is a general species specific course that includes both a didactic and laboratory portion.

Time: 0830-1300

Schedule:

29 May 1998

28 August 1998

5 November 1998

Swine Class

General Information – The Swine Class is a general species specific course that includes both a didactic and laboratory portion.

Time: 0830-1300

Schedule:

6 March 1998

22 May 1998

18 September 1998

12 November 1998

Aseptic and Sterile Techniques Class

General Information – The Aseptic and Sterile Techniques workshop reviews the principles of aseptic and sterile techniques required by federal law to support rodent survival surgeries. Survival surgeries involving animals (other than rodents) must be performed in a dedicated surgery. The class includes both didactic and laboratory sections. Live animals are not used. The workshop is open to investigators and technicians.

Time: 0830-1230

Schedule:

10 April 1998

22 July 1998

30 October 1998

10 December 1998

Introduction to Laboratory Animals Workshop

General Information – The Introduction to Laboratory Animals Workshop is offered for high school, college, and other summer hire/contract personnel, is general in scope, and attendees do not need technician level skills to participate. In previous years, this course has been taken by students working in both animal and nonanimal use laboratories (students from the nonanimal laboratories took the course for general information and education).

The course provides students with a broad overview of laboratory animal care and use policies, practices and procedures, and nonhuman primate safety, and includes a tour of the animal facilities. The “hands-on” portion involves instruction in basic handling and care of rodents and rabbits. This course does not include instruction in research techniques and DoD workshop certificates are not given to attendees. Students actively involved in research must also take the regular DoD workshop for that species.

There is no “hands-on” portion involving nonhuman primates associated with this workshop. Students who have any occasion to enter nonhuman primate rooms or work with nonhuman primates must have clearance from the Director, Division of Veterinary Medicine, and must take the regular DoD Nonhuman Primate Laboratory Animal Workshop.

Time and Schedule:

30 June 1998

0830-1200

7 July 1998

0830-1200

8 July 1998

1230-1600

Appendix L

IACUC Training and Information

Appendix L

IACUC Training and Information

Nonaffiliated IACUC Member Training Recommendations

The following are some example topics and resources which would fulfill the congressionally mandated 8-hour training requirement for any new nonaffiliated IACUC members. This is just one example of a program that would fulfill this training.

Topics:

1. Humane Care and Ethics Issues Dealing with Animal Use (This block should be no longer than 4 hours)
2. Regulatory Responsibilities and Protocol Review Techniques (This block should be no longer than 4 hours)
3. Facility Familiarization Tour
4. Basic Husbandry and Techniques of Laboratory Animals
5. Documentation of Training

Resources:

- Video (40 minutes) "IACUC Functions and the Humane Care and Use of Animals" available from the Laboratory Animal Training Association (LATA)
- Questions and answers with the attending veterinarian
- USAMRIID slide set (~200 slides covering Surgery, Euthanasia, Ethics, Pain and Distress)
- Education and Training in the Care and Use of Laboratory Animals (National Academy Press, 1991)
- Overview of DoD protocol format with the attending veterinarian
- Laboratory animal protocol review articles (available from the editor as a bound notebook with 2 years of reviews)
- USAMRIID slide set covering responsibilities, laws and regulations (~100 slides)
- Attending veterinarian, facility manager, IACUC members
- LATA video tapes and script
- ACLAM slide sets with audio cassettes
- USAMRIID slide set
- Each institute will develop a checklist and sign-in logo to verify training received

Additionally, we recommend the individual institute supplement in-house training programs by sending IACUC members to outside meetings such as Public Responsibility in Medicine and Research (PRIM&R)/Applied Research Ethics, National Association (ARENA) and American Association of Laboratory Animal Science.

Examples of Training and Information Provided to IACUC Members

- Provision of guide books and policy documents such as the: OPRR Institutional Animal Care and Use Guidebook; NIH Publication 85-23, Guide for the Care and Use of Laboratory Animals; the Public Health Service Policy on Humane Care and Use of Laboratory Animals; and the Animal Welfare Act
- Provision of DoD documents and institutional manuals on animal care and use regulations and policy
- Provision of subscriptions to the journal, "Lab Animal" and the newsletter of the National Association for Biomedical Research
- Provision of videotapes and slide sets
- Instruction of AAALAC program accreditation requirements
- One-on-one briefings by attending veterinarians
- Provision of instruction regarding animal use ethics in workshops and/or research training courses
- Funding of attendance at workshops by Scientists Center for Animal Welfare
- Funding of attendance at the PRIM&R conference and/or that of the ARENA
- Provision of monthly, continuing education training materials to each member
- Regular committee discussions of journal and newsletter articles
- Provision of membership in the American Association of Laboratory Animal Science
- Provision of course on animal use and handling commercial firm (Renaissance Research Associates, Inc), and/or Animal Welfare Information Center training
- Maintenance of animal use resource libraries variously containing regulatory documents, protocols, course materials, journals, videos, slide sets, and literature on animal care and use
- Provision of a seminar on the role of nonaffiliated and nonscientist IACUC members

Appendix M

Status of AAALAC Accreditation of DoD Animal Care and Use Facilities

Appendix M

Status of AAALAC Accreditation of DoD Animal Care and Use Facilities

I U.S. DoD Programs Accredited by AAALAC:

I.1 OSD Components:

- Armed Forces Institute of Pathology, Washington, DC
- Armed Forces Radiobiology Research Institute, Bethesda, MD
- Uniformed Services University of the Health Sciences, Bethesda, MD

I.2 U.S. Army:

- U.S. Army Research Institute of Environmental Medicine, Natick, MA
- U.S. Army Medical Research Institute of Chemical Defense, Aberdeen Proving Ground, MD
- U.S. Army Medical Research Institute of Infectious Diseases, Fort Detrick, MD
- U.S. Army Center for Environmental Health Research, Fort Detrick, MD
- U.S. Army Edgewood Chemical Biological Center, Edgewood, Aberdeen Proving Ground, MD
- William Beaumont Army Medical Center, El Paso, TX
- Tripler Army Medical Center, Tripler Army Medical Command, Honolulu, HI
- Madigan Army Medical Center, Tacoma, WA
- U.S. Army Center for Health Promotion and Preventive Medicine, Aberdeen Proving Ground, MD
- U.S. Army John F. Kennedy Special Warfare Center , Fort Bragg, Fayetteville, NC
- Walter Reed Army Institute of Research, Washington, DC
- Brooke Army Medical Center, Fort Sam Houston, TX
- U.S. Army Medical Department Center and School, Fort Sam Houston, TX
- Dwight David Eisenhower Medical Center, Fort Gordon, GA
- U.S. Army Dugway Proving Ground, Dugway, UT
- U.S. Army Institute of Surgical Research, Fort Sam Houston, TX

I.3 U.S. Navy:

- Naval Dental Research Institute, Naval Training Center, Great Lakes, IL
- Naval Medical Center, Clinical Investigation Program, San Diego, CA
- Naval Medical Center, Clinical Investigation and Research, Portsmouth, VA
- Naval Medical Research Institute, Bethesda, MD
- Space and Naval Warfare Systems Center, San Diego, CA

I.4 U.S. Air Force:

- Air Force Research Laboratory, Wright-Patterson Air Force Base, OH
- Air Force Research Laboratory, Brooks Air Force Base, TX
- Keesler Medical Center, 81st Medical Group, Keesler Air Force Base, MS
- Wilford Hall Medical Center, 59th Medical Wing, Lackland Air Force Base, TX
- David Grant Medical Center, 60th Medical Group, Travis Air Force Base, CA
- U.S. Air Force Academy, Colorado Springs, CO

II Overseas Programs Accredited by AAALAC:

- Naval Medical Research Institute Detachment, Lima, Peru
- Naval Medical Research Unit #2, Jakarta, Indonesia
- Naval Medical Research Unit #3, Cairo, Egypt

III DoD Programs Not AAALAC Accredited:

- Armed Forces Research Institute of Medical Sciences, Bangkok, Thailand
- U.S. Navy Camp Lejeune Marine Corps Base, NC
- U.S. Army Landstuhl Regional Medical Center, Germany

Appendix N

Animal Use Categories

Appendix N

Animal Use Categories

MEDICAL (M)

M1: Military Dentistry

Includes studies in the areas of:

- dental disease and management of dental emergencies
- testing medical devices for maxillofacial injury
- testing materials for maxillofacial injury
- surgical management of maxillofacial injury

M2: Infectious Diseases

Includes studies in the areas of:

- emerging infectious diseases of military importance
- vaccine development for prevention of bacterial sepsis and septic shock
- shigella vaccines
- malaria vaccines
- gonococcal peptide vaccine
- enterotoxigenic *E. coli* (ETEC) vaccine
- rickettsial diseases
- group A streptococcal vaccines
- polyvalent meningococcal vaccine
- prevention of *Campylobacter* diarrheal disease
- hepatitis virus vaccines
- establishment of diagnostic tests for infectious disease agents
- diagnosis of leishmaniasis
- development of drug therapies for infectious disease agents
- dengue virus vaccines
- viral hemorrhagic fever and encephalitis prevention and countermeasures
- identification and control of insect vectors of infectious diseases
- prevention of military HIV infection

M3: Medical Chemical Defense

Includes studies in the development of:

- medical countermeasures for vesicant agents
- medical pretreatment for cyanide
- prophylactic therapeutics for chemical agents
- reactive topical skin protectant
- medical countermeasures for respiratory agents
- chemical casualty management strategies and treatments

M4: Medical Biological Defense

Includes studies in the development of medical countermeasures for:

- *Yersinia pestis*
- brucellosis
- anthrax
- *Clostridium perfringens*
- Q-fever
- *Francisella tularensis*
- encephalomyelitis viruses
- variola
- Filoviridae
- physiologically active compounds
- sodium channel neurotoxins
- ricin
- staphylococcal enterotoxin B
- botulinum toxin
- venoms

M5: Human Systems Technology

Includes studies on:

- bioeffects of lasers
- laser impacts on performance
- treatment of laser-induced injury
- development of predictive models for a non-auditory exposure standard for blast overpressure

- development of occupational health protection criteria and exposure assessment technologies for toxic hazards arising from weapon systems and combat operations
- vibration
- bioeffects of electromagnetic radiation
- development of countermeasures for the effects of operational stress on military performance
- environmental injury
- development of methods, criteria, and predictive models for the risk of pulmonary injury in defeated armor scenarios

M6: Combat Casualty Care

Includes studies in:

- blood loss
- resuscitation
- secondary damage after hemorrhage
- soft tissue injury
- musculoskeletal injury
- combat stress injury
- burn injury
- anesthetics
- delivery systems

M7: Ionizing Radiation

Includes studies on:

- development of radioprotective compounds
- therapies for radiation-induced pathology
- bioeffects of ionizing radiation
- psychomotor effects of ionizing radiation
- mechanisms of radiation-induced pathophysiology

M8: Other Medical RDT&E

Includes studies in the areas of:

- breast cancer research
- neurofibromatosis research
- Gulf War illnesses
- laser research
- toxicology
- zoonosis
- free electron laser
- Defense Women's Health Research
- occupational medicine
- osteoporosis

- vectorborne diseases
- prostate cancer
- environmental safety
- disaster relief and emergency medical services

NON-MEDICAL (N)

N1: Physical Protection

As previously indicated, excludes reporting military working animals and includes:

- developing hearing protection criteria
- mechanisms of and protection from military acoustic hazards
- ocular effects and performance of eye protective devices

N2: Physical Detection

Includes studies in the development of:

- biosensors
- chemical detection devices
- the Chemical Biological Mass Spectrometer (CBMS) detector
- auditory detection thresholds in marine mammals
- models of dolphin echolocation
- detection of biological warfare agents

N3: Offensive Weapons Testing

No studies were performed in this category in FY98.

N4: Other Non-Medical RDT&E

Includes studies in the areas of:

- toxicology
- marine biology
- biorobotics
- biosonar
- learning and memory physiology
- environmental research
- biological sensors
- jet lag and wakefulness
- solvent and metal toxicity
- neuroscience
- bionavigation

CLINICAL INVESTIGATIONS (C):

C1: Clinical Medicine

Research conducted includes a wide variety of clinical medical diseases/conditions that are not necessarily unique to the military.

Includes studies in the areas of:

- burn treatment
- prophylaxis against toxic chemicals
- wound healing
- preservation of tissue sample morphology
- differentiation of brain tumors
- substances promoting repair of sound-sensing cells
- regulation of tracheal mucin secretion by retinoic acid
- breast cancer research
- mechanisms and treatment of renal pathophysiology
- effects of tumor necrosis factor on gonadotrophic activity
- treatment of immune-mediated hearing loss
- mechanisms of lung growth and compensation following injury
- testing of hepatitis-E vaccines

C2: Clinical Surgery

Includes studies in the areas of:

- adverse effects on wound healing of post-surgical treatments
- development of synthetic materials for surgical closures
- topical stimulants of skin healing following biopsies
- techniques of fiberoptic bronchoscopy
- laparoscopic cholecystectomy
- biomechanical and histological effects of artificial implants
- identification and development of improved implant materials
- evaluation of new techniques to remove seminal vesicle cysts
- electrohydraulic lithotripsy

C3: Other Clinical Investigations

- medical skills training

TRAINING AND INSTRUCTIONAL (T):

T1: Training, Education, and/or Instruction for Personnel

Types of training include:

- animal technician training
- training of special forces medics
- investigator training in proper techniques used with animals
- physician training in medical or surgical procedures, etc.

The training locations included DoD laboratories or medical centers. Does not include experimental or research-related work.

T2: Other Training/Instruction

No studies were performed in this category in FY98.

ADJUNCTS AND ALTERNATIVES TO ANIMAL STUDIES (A):

A1: Adjuncts to Animal Use Research

Addresses those studies and uses that focus specifically on animal husbandry and care issues, and not directly on human medical, non-medical, or training issues.

A2: Alternatives to Animal Investigation

Includes studies involving the use of animals that are designed to address directly and specifically issues of replacement, reduction, or refinement options for which animals are currently used; this classification does not include studies that are specifically directed at military RDT&E, clinical studies, or training requirements that may employ the animal alternatives of replacement, reduction, or refinement in the performance of the required protocols.

A3: Other Alternatives/Adjuncts

No studies were performed in this category in FY98.

**CLASSIFIED SECRET OR ABOVE
STUDIES (S):**

**S: Animals in Studies Classified SECRET or
Above**

Includes studies in which the information concerning the study may not be released for public knowledge because of the impact on national security.

ANIMAL BREEDING STOCK (B):

B: Animals Maintained for Breeding

Includes:

- animals maintained at the facility or supported through contract funds for breeding purposes to supply offspring to be used in animal-based research for particular work units or protocols

- breeding animals and offspring not assigned to specific work units or protocols

OTHER ANIMAL USE CATEGORIES (O):

O: Other Animal Use Purposes

Includes:

- animals awaiting assignment to protocols
- environmental monitoring
- quality assurance
- behavioral studies
- biomonitoring
- bioassays
- source of red blood cells
- FDA-sponsored clinical studies
- Pro-Naron-sponsored clinical studies

Appendix O

Summary of Animal Use Data by Category

Appendix O

Summary of Animal Use Data by Category

(M1) Military Dentistry

<i>Animals Reported</i>	<i>Animals Used</i>
Mouse	150
Total	150

(M2) Infectious Diseases

<i>Animals Reported</i>	<i>Animals Used</i>
Bird	154
Calf	3
Chicken	620
Crane	16
Dog	99
Gerbil	30
Goose	9
Guinea Pig	773
Hamster	1,754
Mouse	66,776
Nonhuman Primate	769
Rabbit	277
Rat	1,130
Sheep	20
Total	72,430

(M3) Medical Chemical Defense

<i>Animals Reported</i>	<i>Animals Used</i>
Frog	7
Guinea Pig	2,948
Mouse	32,076
Nonhuman Primate	118
Pig/Swine	94
Rabbit	776
Rat	2,100
Total	38,119

(M4) Medical Biological Defense

<i>Animals Reported</i>	<i>Animals Used</i>
Burro	2
Gerbil	11
Goat	6
Goose	8
Guinea Pig	7,318
Hamster	430
Horse	3
Mouse	37,885
Nonhuman Primate	297
Rabbit	207
Rat	203
Sheep	33
Total	46,403

(M5) Human Systems Technology

<i>Animals Reported</i>	<i>Animals Used</i>
Frog	422
Hamster	660
Mouse	2,502
Nonhuman Primate	63
Pig/Swine	264
Rabbit	237
Rat	2,869
Sheep	12
Toad	58
Total	7,087

(M6) Combat Casualty Care

<i>Animals Reported</i>	<i>Animals Used</i>
Dog	1
Goat	85
Guinea Pig	39
Mouse	1,525
Nonhuman Primates	61
Pig/Swine	714
Rabbit	815
Rat	4,835
Sheep	31
Total	8,106

(M7) Ionizing Radiation

<i>Animals Reported</i>	<i>Animals Used</i>
Dog	41
Guinea Pig	178
Mouse	5,716
Rat	851
Total	6,786

(M8) Other Medical RDT&E

<i>Animals Reported</i>	<i>Animals Used</i>
Cat	1
Chicken	188
Chinchilla	31
Chipmunk	7
Dog	18
Frog	547
Guinea Pig	38
Lemming	1
Minnow	240
Mouse	51,249
Nonhuman Primate	59
Pig/Swine	7
Rabbit	172
Rat	6,917
Sheep	13
Shrew	36
Squirrel	10
Vole	83
Wren	1
Total	59,618

(N1) Physical Protection

<i>Animals Reported</i>	<i>Animals Used</i>
Hamster	13
Mouse	809
Nonhuman Primate	133
Rabbit	17
Rat	576
Total	1,548

(N2) Physical Protection

<i>Animals Reported</i>	<i>Animals Used</i>
Bat	3
Big Brown Bat	33
Chicken	20
Goat	11
Mouse	835
Rabbit	128
Rat	88
Snake	14
Total	1,132

(N3) Offensive Weapons Testing

<i>Animals Reported</i>	<i>Animals Used</i>
Total	0

(N4) Other Non-Medical RDT&E

<i>Animals Reported</i>	<i>Animals Used</i>
Boxfish	25
California Sea Lion	2
Cat	20
Degu	25
Fathead Minnow	720
Ferret	14
Fish	1,000
Gerbil	35
Guinea Pig	75
Hamster	467
Iguana	158
Mouse	2,933
Nonhuman Primate	11
North Elephant Seal	1
Opossum	10
Pacific Harbor Seal	1
Pig/Swine	18
Pigeon	24
Rabbit	381
Rat	3,474
Sea Turtle	1
Snake	41
Squirrel	22
Sunfish	6
Vole	290
Total	9,754

(C1) Clinical Medicine

<i>Animals Reported</i>	<i>Animals Used</i>
Chinchilla	106
Dog	19
Ferret	132
Goat	7
Guinea Pig	88
Mouse	5,648
Nonhuman Primate	84
Pig/Swine	244
Rabbit	481
Rat	3,593
Sheep	5
Total	10,407

(C2) Clinical Surgery

<i>Animals Reported</i>	<i>Animals Used</i>
Dog	98
Ferret	5
Goat	56
Pig/Swine	393
Rabbit	131
Rat	210
Sheep	4
Total	897

(C3) Other Clinical Investigations

<i>Animals Reported</i>	<i>Animals Used</i>
Pig/Swine	35
Rabbit	4
Rat	77
Total	116

**(T1) Training, Education, and/or
Instruction of Personnel**

<i>Animals Reported</i>	<i>Animals Used</i>
Bullfrog	28
Cat	15
Chicken	25
Dog	29
Ferret	100
Gerbil	4
Goat	2,074
Grass Frog	36
Guinea Pig	90
Hamster	8
Mouse	275
Nonhuman Primate	15
Pig/Swine	791
Rabbit	84
Rat	1,202
Sheep	32
Total	4,808

(T2) Other Training/Instruction

<i>Animals Reported</i>	<i>Animals Used</i>
Nonhuman Primate	20
Total	20

(A1) Adjuncts to Animal Use Research

<i>Animals Reported</i>	<i>Animals Used</i>
Dolphin	33
False Killer Whale	1
Guinea Pig	10
Mouse	409
Nonhuman Primate	4
Rabbit	18
Rat	11
Sea Lion	2
White Whale	2
Total	490

(A2) Alternatives to Animal Investigation

<i>Animals Reported</i>	<i>Animals Used</i>
African Clawed Frog	270
Bluegill Sunfish	45
Dolphin	4
Japanese Medaka	16,000
Jird	45
Total	16,364

(A3) Other Alternatives to Animal Investigation

<i>Animals Reported</i>	<i>Animals Used</i>
Total	0

(S) Classified Secret or Above

<i>Animals Reported</i>	<i>Animals Used</i>
Goat	32
Nonhuman Primate	50
Pig/Swine	21
Rat	171
Total	274

(B) Breeding Stock

<i>Animals Reported</i>	<i>Animals Used</i>
Mouse	3,862
Total	3,862

(O) Other Animal Use Categories

<i>Animals Reported</i>	<i>Animals Used</i>
African Clawed Frog	272
Bluegill Sunfish	2,732
Dog	4
Ferret	12
Killifish	72
Mouse	51
Rabbit	3
Rat	34
Total	3,180

GRAND TOTAL ANIMAL USE/RESEARCH

291,551

Appendix P

Alternatives

Appendix P

Alternatives

Replacement - The replacement alternative addresses supplanting animal use with non-living systems, analytical assays, cell-culture systems, and with animals that are lower on the phylogenetic scale. Additionally, human subjects are used when experimental drugs and other procedures progress to human trials. Such trials are conducted in accordance with Title 32, U.S. Code of Federal Regulations, Section 219, "Protection of Human Subjects in DoD-Sponsored Research."

Alternative Tissue/Organ Collection Techniques

- In medical free electron laser research, human skin obtained as excess surgical tissue is replacing the use of animal skin.
- Bovine cortical bones obtained from a slaughter house are used instead of live laboratory animal cortical bone specimens.
- In vitro models in the Tri-Service Cataract Course and using harvested rabbit eyes from a commercial source have replaced the use of pigs.

Inanimate Training Model Substitution

- During microvascular surgery training, suturing is initially taught on inanimate practice cards, replacing some animals used.
- In clinical assistant trauma training, the goat has been replaced with trauma mannequins.
- In OB/GYN microsurgical training, inanimate materials are used for the first 2 of 4 sessions (intro instrumentation and suture orientation and handling) instead of animals.
- Mannequins and other training aids are used to replace pigs in operational and emergency medicine training courses for as much of the course as possible.
- Mannequins and other training aids are used to replace animals for as much of the course as possible.

Non-Mammalian Species or Species Lower on Phylogenetic Scale

- Studies on cellular mechanism of mucin secretion are performed on rat and rabbit tracheal cultures instead of nonhuman primates, cats, or dogs.
- The use of BALB/c mice replaces guinea pigs and nonhuman primates in Ebola research.
- Vaccine formulations are screened in a mouse model before moving into testing in nonhuman primates.
- Guinea pigs are used as an alternative to ongoing anthrax vaccine protection studies in rabbits and nonhuman primates for anthrax vaccine potency.

- Mice replace nonhuman primates in research to determine protective antigenic epitopes important to vaccine development.
- Filovirus vaccine candidates were prescreened in a guinea pig model system and shown to be efficacious, replacing the use of nonhuman primates.
- The use of guinea pigs replaces primates in chlamydia studies.
- Rats replace sheep in studies on the regulation of perinatal hepatic glutamate flux.
- Special Projects in Behavioral Pharmacology are now being performed on rats instead of baboons.
- The use of mice replaces the use of nonhuman primates in studying the pathological reactions of toxic shock syndrome toxin 1 and development of a prototype vaccine and treatment agents.

Other Species Replace Companion Animals

- The rabbit model replaces the use of dogs in platelet studies.
- In emergency skills training, pigs have replaced the use of dogs.
- In the bronchoesophagology course, ferrets replaced cats.

Replacement Using In Vitro Cell Cultures

- Neutralization assays are conducted in cell culture assays to select only antibodies that have a good chance of protecting from infection in Hantavirus studies.
- Neutralization and ELISA assays are conducted in cell culture assays to select the most promising genes for vaccine efficacy in filoviruses, Hantaviruses, tick-borne encephalitis viruses and vaccinia virus studies.
- Cell culture assays for determining the neutralization titers of sera replace the requirement for animals.
- In Alphavirus vaccine testing, cell culture assays for determining the neutralization titers of sera replace the requirement for animals.
- Susceptible cell culture lines are determined and subsequently used for virus propagation after initial isolation in mice.
- Antiviral drugs are initially tested in vitro for the ability to inhibit Marburg and Ebola virus replication. Drugs that do not show activity in vitro are not tested in animals.
- In screening experiments for optimizing radiation dose response, assays are carried out on cell lines instead of animals.
- Cultured rat cells are used instead of animals to test for various toxicity endpoints in solvent and metal toxicity studies.

Reduction - Decreasing the number of animals used through the use of statistical or innovative design strategies, while preserving the scientific integrity of the biological model, is a major emphasis of the reduction alternative to animal use.

Alternative Tissue/Organ Collection Techniques

- Substitution of living tissue with cadaveric tissue homografts reduces the number of pigs required for sources of tracheal tissue.
- Bovine brain obtained from a local slaughter house is used to characterize brain receptors.

Enhanced Experimental Design

- Improvement in methods to maintain colonies of chiggers reduces the number of animals required.
- Use of Thompson's Moving Average Interpolation requires fewer animals than the more stringent Probit analysis, yet permits a statistically valid estimate of lethal dose.
- In wound healing studies, 10 different keratinocyte/fibroblast constructs are studied in each animal.

Non-Mammalian Species or Species Lower on Phylogenetic Scale

- The use of rabbits in anthrax vaccination studies reduces the number of nonhuman primates that would otherwise have to be used in these efficacy studies.
- The use of guinea pigs to study Ebola virus pathogenesis reduces the number of nonhuman primate studies.
- The use of rats for screening of streptococcal hypertensive compounds greatly reduces the number of lambs required.
- Evaluation of the activity of antiviral compounds against Ebola virus disease in mice serves as a screening method to reduce the number of compounds requiring testing in nonhuman primates.
- Dengue vaccine candidates are tested in mice prior to their being tested in monkeys; candidates with sub-optimal efficacy in mice are eliminated.
- Preliminary screening of all malaria vaccines in guinea pig toxicity and mouse immunogenicity models reduces the number of formulations tested in monkeys.
- Preliminary testing of antiviral compounds in the cowpox mouse model reduces the number of compounds that require evaluation in nonhuman primates.
- The use of guinea pigs in this study reduces the number of rabbits and nonhuman primates that would have to be used to investigate *B. anthracis* strain differences
- The use of guinea pigs to study Ebola virus pathogenesis helps to reduce the number of nonhuman primate studies needed to gain a full understanding of Ebola infection in man.

Substitution of Computer Simulation Models or Other Technologies

- Utilizing inanimate objects, videos, and multiple procedures on one animal reduced the use of animals in the basic microsurgery course.
- In the characterization of recombinant bacterial superantigen vaccines, computer modeling and in vitro testing are used to decrease the number of animals required.
- The number of animals utilized was reduced through the use of computer modeling of potential peptide antigens to determine if the peptide has the same conformation as the analogous sequence in the native protein.
- In studying the effect of laser exposures off the visual axis, a mathematical model developed to predict the effects of off-axis exposure reduced the number of animals needed.
- Students used model eyes when using lasers for the first time, reducing the number of animals used.

Utilization of Alternative Biological Testing Method

- To minimize the number of animals needed, the same frozen tissue was used on multiple growth factors and receptors.
- By allowing the use of myocytes from a single animal in multiple experiments, the number of animals is reduced.
- By using weight bearing and non-weight bearing surfaces in the same joint, the total number of goats needed for the study of implants was decreased by 50%.
- In repellent studies, the in vitro test systems reduces the number of animals used by identifying potential repellents and providing an estimate of optimum concentration to repel a given insect/arthropod population.
- An in vitro system to supplement ascites production of monoclonal antibodies reduced the number of mice used.
- Mutant bacteria are tested first in a human monocyte cell culture system for infectivity, growth and survival before being tested in animals. Only those mutants that are clearly attenuated under in vivo conditions are tested further in animals.
- In vitro susceptibility testing is used to screen test antimalarial compounds, thus reducing the number of animals required for in vivo drug assessment.
- The sequential dosing method results in a reduction of the number of animals required.
- In monkeypox and Ebola Zaire research, the use of a cell culture assay for initial determination of antiviral activity greatly reduces (90%) the number of compounds requiring testing in primates.
- The viral infection process in the mosquito was “partitioned,” ensuring that every mosquito disseminates infection prior to feeding on an animal; this decreases the number of animals needed.

- By combining the lethality/safety study with the efficacy study, a reduction of the number of animals used is realized as compared to running the studies separately.
- A modified staircase method was used to determine relative susceptibility values, which reduces the number of animals needed.
- Combining safety testing, immunogenicity evaluation, and challenge studies in the same animals, and the use of challenge criteria to minimize the number of monkeys used and/or challenged with SEB toxin reduce the number of monkeys required.
- Separate studies using in vitro methods are conducted as a primary screen to the in vivo neuroprotection studies as an alternative approach to minimize, but not substitute for in vivo experiments.
- In radiation injury studies, the use of in vitro cell cultures reduced the number of animals needed.
- Ceriodaphnia and MICROTOX assays are used to determine toxicity estimates and reduce the number of vertebrates required to perform survival and growth tests.

Refinement - The refinement alternative for animal use addresses the need to ensure that the maximum humane use of each animal is obtained through proper protocol design and efficient utilization of animals, or through the modification of the experimental design to reduce the ethical cost associated with the study.

Environmental Enrichment and Improved Animal Handling

- In training support personnel and investigators, technique videos and non-animal models are used prior to animal use.
- Rabbits are provided enrichment toys.
- While in issue pool, animals participate in environmental enrichment program. Examples of enrichment include: food treats, provision of toys, pair housing, and family housing.
- Enrichment activities to reduce goat stress include alpha bucks to maintain social order, and climbing, rolling, and reaching toys.
- The monkeys were pair caged to minimized distress, unless otherwise justified by the facility veterinarian. Various enrichment devices were utilized, e.g., perches, swings, kong-toys, balls, puzzles, foraging boards, and mirrors.

Reduce Distress

- To minimize stress or pain to the animal during handling while training research personnel, chemical restraint is used. The animals experience little to no pain or discomfort because the first injection given is the anesthetic.
- Antemortem biosamples, e.g., blood, are obtained while the animals are anesthetized.

- Improved postoperative care training of veterinary staff significantly increased postoperative survival while reducing recovery distress.
- Sheep were conditioned to the exercise requirements of the protocol in advance of surgical device implantation.
- Although mosquito feeding is not considered painful, isoflurane is used as a short-term restraining agent to reduce handling stress.
- The use of continuous monitoring of physiological parameters by telemetry in an unrestrained Ebola monkey and cynomolgus monkeypox model reduces distress.
- Use of light anesthesia to mice before examining them for ticks reduces their stress; mice serve as their own controls.
- Severely moribund mice are euthanized as soon as they develop hind limb paralysis, a better endpoint/indicator of morbidity.
- Close monitoring for parasitemia and prompt antimalarial treatment after mosquito feeding ensures that minimal discomfort is experienced from the infection. Anesthesia is used to eliminate distress during mosquito feeding.
- Use of implantable temperature transponders is less distressful than invasive rectal thermometers.
- Visual discrimination data are collected continuously throughout light and dark cycles from rats housed in their “home” conditioning chambers. This approach to behavioral assessment maximizes data obtained from each subject while minimizing handling.
- Use of fewer sarcoma cells per mouse decreases the likelihood of solid tumor production in adult mice, thereby reducing the amount of distress in these animals.

Reduce Pain

- In burn research, Telazol anesthesia replaced ketamine, resulting in significantly less volume intramuscularly, thereby lessening momentary pain/distress as well as long-term muscle damage.
- Twenty-four hour monitoring is performed for the initial 2 days post-operatively to ensure minimal pain and distress from surgical intervention.
- Several of the mosquito colonies were adapted to membrane feeding, thereby reducing pain.
- The use of post-surgical analgesics reduces pain.
- Mannequins are used initially to teach the intubation technique, thereby reducing the potential trauma induced in the live animal.
- Limitations are set on the number of intubations per cat per session to limit possible pharyngeal trauma.

- Animal pain is minimized by the use of the most current analgesics. Opioids, being ineffective analgesics on goats, are being replaced with alpha agonists to include medetomidine.
- By euthanizing the animals while still under anesthesia and not allowing them to recover to evaluate the success of the procedures, the chance of subjecting the animals to severe pain has been eliminated.
- Surface electrodes are non-invasive and more humane.

Utilization of Alternative Biological Testing Methods

- In spinal cord research, the post-surgical survival time was reduced from 90 days to 30 days.
- By eliminating the use of flow probes around the renal arteries, the chance of a problem of hemorrhage or occlusion that would result in loss of data from that animal was reduced.
- In Scoliosis treatment research modifying the stapling technique in this protocol, less of the vertebral body is removed, thereby causing less trauma to the goat in the area of the spinal cord.
- The use of radiotelemetry reduces stress to the animal and improves the quality of the data obtained.

Appendix Q

Food and Drug Administration Group Recognition Award



**Food and
Drug
Administration**

**Group
Recognition
Award**

PRESENTED TO
Harry Salen

*as a member of Interagency Regulatory Alternatives
Group (IRAG)*

*For outstanding contribution in facilitating the reduction, refinement and replacement
of animal testing, and advancing the development of non-whole animal alternative
methods.*

May 9, 1997

DATE

Michael A. Friedman

LEAD DEPUTY COMMISSIONER
FOOD AND DRUG ADMINISTRATION